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BOTANICAL SCIENCES

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VOL. 26, SEC. C. DECEMBER, 1948 NUMBER 6

A COMPARATIVE STUDY OF THE ACTIVITY OF FIFTY ANTIBIOTIC ACTINOMYCETES AGAINST A VARIETY OF SOIL BACTERIA[1]

BY G. B. LANDERKIN[2] AND A. G. LOCHHEAD[3]

Abstract

Fifty actinomycetes, isolated from a variety of soils on the basis of their antibiotic activity against *Escherichia coli*, were tested against different soil bacteria. Wide variations were noted in susceptibility of the test organisms to the antagonists. Actinomycetes with the most intense antibiotic activity were able to act upon the greatest number of bacterial species.

In connection with an investigation of the occurrence of antibiotic actinomycetes in soil and their relation to specific soil conditions, a study was made of the action of 50 strains, isolated from Canadian soils from widely separated regions, against a variety of soil bacteria.

The antibiotic activities of actinomycetes and aspects of the physiology of this group of organisms have been summarized by Waksman (6, 7). Mention might be made of the work of Nakhimovskaia (3) who studied antagonistic actinomycetes isolated from a variety of Russian soils that were active against Gram-positive bacteria, though not against Gram-negative bacteria or fungi. Waksman *et al.* (8) found antagonistic forms widely distributed in soils and composts, strains possessing some activity comprising 43% of those isolated. Nickell and Burkholder (4) demonstrated the inhibition of *Azotobacter vinelandii* by 25 cultures of actinomycetes isolated from soil and found the antagonistic action still evident in the presence of added soil, fertilizers, and crop residues. More recently Perrault (5) reported the isolation, from decaying potatoes, of a number of organisms, among which were four strains of actinomycetes that showed antibiotic properties against *Corynebacterium sepedonicum*. In a recent study of the action of actinomycetes against *Rhizobium*, Fogle and Allen (1) noted considerable difference in susceptibility shown by different species of this genus towards the action of various strains of actinomycetes.

Experimental

Thirty-six cultures of actinomycetes were obtained by the direct method for isolating antagonistic forms described by Landerkin (2). Plates of Conn's

[1] *Manuscript received August 5, 1948.*

Contribution No. 263 (Journal Series) from the Division of Bacteriology and Dairy Research, Science Service, Department of Agriculture, Ottawa.

[2] *Associate Bacteriologist.*

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[The October issue of Section C (Can. J. Research, C, 26: 461-500. 1948) was issued November 8, 1948.]

asparagine agar were prepared from appropriate soil dilutions and incubated for five days at 26° C., at which time those plates showing 50 to 150 colonies were flooded with 3.5 ml. of yeast beef agar (Difco) seeded with 1% of a 24-hr. culture of *Escherichia coli* grown in yeast beef broth at 37° C. The flooded plates were incubated an additional 24 hr. at 37° C., the Petri dish cover being replaced by a sterile porous porcelain cover, and were then examined for zones of inhibition. Actinomycete colonies antagonistic to *E. coli* were surrounded by a clear zone of agar. Strains showing most pronounced inhibition were inoculated to slants of asparagine agar, and after growth and replating on the same medium, were reisolated in pure culture.

The remaining 24 cultures were selected from a group of 700 strains of actinomycetes isolated from soil,* and chosen on the basis of their antibiotic activity against *E. coli* when tested individually.

In order to determine the antagonistic effects of growth of the selected actinomycetes upon a series of soil organisms a spot inoculation procedure was employed. Standard Petri plates were poured with 21 ml. of asparagine agar and held for 24 to 48 hr. at room temperature for hardening and evaporation of excess surface moisture. Each plate was then spot inoculated with five different actinomycetes by touching a 3-mm. loop of a spore suspension to the surface of the agar in clockwise manner equidistant from each other, using a cardboard guide below the plate for accuracy. The plates were then incubated at 26° C. for a period of four to six, normally five, days for development of colonies prior to testing.

Actinomycete spore suspensions were prepared by scraping the surface of 8- to 12-day-old asparagine agar cultures into approximately 10 ml. of a freshly prepared sterile solution of Tween "20" diluted to 1 : 5000 with distilled water. In this manner spores were suspended without inhibition of further growth or subsequent mutation.

To obtain a profile of the relationship of the 50 antibiotic actinomycetes towards a series of organisms the following species were chosen:

Escherichia coli (117), *Erwinia carotovora* (529), *Xanthomonas campestris* (589), *Xanthomonas phaseoli* (593), *Bacterium globiforme* (425); all grown on yeast beef agar.

Azotobacter vinelandii (534), *Azotobacter chroococcum* (536), *Rhizobium meliloti* (309), *Rhizobium japonicum* (462), *Rhizobium trifolii* (311), *Corynebacterium sepedonicum* (610 and 611); all grown on mannitol agar of the following composition:— mannitol, 10 gm.; dipotassium hydrogen phosphate, 0.5 gm.; magnesium sulphate septahydrate, 0.2 gm.; sodium chloride, 0.1 gm.; calcium chloride, 0.1 gm.; yeast extract, 1.0 gm.; mesquite gum hydrolyzate, 5 ml. (equivalent to 2 gm. mesquite gum); agar, 15 gm. with tap water to make one liter.

Actinomyces scabies (597 and 598), both cultivated on Conn's asparagine agar.

* *We are indebted to Mr. J. W. Rouatt for supplying these isolates for testing.*

The susceptibility of the above cultures, with the exception of *A. scabies*, to the antagonistic actinomycetes was studied by the flood plate technique. Suspensions of each organism were prepared in physiological saline from 4- to 6-day agar slants to a turbidity roughly equivalent to that of a 24-hr. broth culture of *E. coli*, and each suspension in turn was used to seed a small flask of cooled medium similar to that used for cultivation at a level of 1 to 1½% depending upon relative turbidity. In the case of *R. japonicum* and *C. sepedonicum* the agar was seeded at a level of 2 to 2½% to compensate for a somewhat slower growth. When the seed agar was prepared, 3.0 ml. was used to flood the surface of each asparagine agar plate having five actinomycete colonies of approximately five days' development. All plates were then returned to 26° C. for further incubation with the exception of those flooded with *E. coli*, which were incubated at 37° C. Cultures of *A. scabies* were tested by the cross streak method using a spore suspension in Tween "20" 1 : 5000 and a 3 mm. loop for cross inoculation of the original streak of selected antibiotic actinomycetes.

Antagonism of the test organism was estimated by measuring the diameter of the zone of inhibition after four days' incubation at 26° C. (with *E. coli* readings were taken after 24 hr. at 37° C.). Irregularities in colony shape as well as small differences in actual diameter of original spot inoculated actinomycetes were sources of error; however, the mean diameter measured to nearest millimeter was taken, since differences between duplicate plates prepared in early trials indicated that the uniformity to be expected allowed for measurement to this tolerance. With *A. scabies* the average of measured distance from growth to growth of streaks was taken as equivalent to the zone of inhibition.

Comparative Antibiotic Effects

The antibiotic action of the 50 strains of actinomycetes on the 12 bacterial species (14 strains) is summarized in Table I. For convenience in presentation the degree of antagonism is indicated by the numbers 1 to 4, depending upon the extent of the zone of inhibition. This 'profile' determination indicates considerable differences in the bacterial susceptibility to the antagonistic organisms. Thus in the case of *C. sepedonicum* inhibitions were mostly at the maximum degree, whereas with most other organisms, there was a wider band of less pronounced response. The similarity of reaction observed with duplicate strains of *C. sepedonicum* and *A. scabies* testifies to the reliability of the method, zone diameters and intensity ratings following like patterns. Whereas the two species of *Azotobacter* showed very similar reactions to the antagonists, the three species of *Rhizobium* showed considerable variation in their susceptibility, particularly as regards intensity ratings.

It will be noted that only 78% of the antagonistic actinomycetes gave measurable zones of inhibition against *E. coli*, though they were selected originally because of antagonism exhibited against this organism. Whether

TABLE I

PROFILE SUMMARY OF 50 ANTIBIOTIC ACTINOMYCETES

Test organism	Actinomycetes inhibiting		Average zone diam.	Degree of antagonism*				
	No.	%		4	3	2	1	0
E. coli (117)	39	78	21.1	16	4	13	6	11
A. vinelandii (534)	36	72	27.1	21	9	3	3	14
A. chroococcum (536)	36	72	25.2	19	8	5	4	14
R. meliloti (309)	38	76	19.2	8	10	16	4	12
R. japonicum (462)	30	60	26.6	19	5	3	3	20
R. trifolii (311)	40	80	23.2	20	4	12	4	10
B. globiforme (425)	47	94	25.1	21	9	14	3	3
E. carotovora (529)	38	76	21.4	11	10	14	3	12
X. campestris (589)	33	66	25.1	18	4	8	3	17
X. phaseoli (593)	36	72	23.7	13	10	11	2	14
C. sepedonicum (610)	47	94	29.7	32	9	6	0	3
C. sepedonicum (611)	48	96	30.2	35	6	7	0	2
A. scabies (597)	44	88	13.7	1	12	16	15	6
A. scabies (598)	44	88	14.0	1	12	16	15	6

* NOTE: *4* = *zone 30 mm. or greater.*
3 = " *20 to 29 mm.*
2 = " *10 to 19 mm.*
1 = " *trace to 9 mm.*
0 = *no inhibition.*

this is to be ascribed to loss of the active strain in purification, mutation on cultivation, or decrease in intensity of antibiotic production within the limits of the test is not known. However, the latter explanation is in most cases most likely, since other species, e.g. *C. sepedonicum* and *B. globiforme*, were inhibited by 96 and 94% respectively of the actinomycetes.

The relative susceptibilities of the 12 test organisms to the 50 selected actinomycetes are shown in Fig. 1, where *E. coli* is taken as the standard reference with a rating of 100 and all other test cultures compared to this culture. Thus *C. sepedonicum* would appear to be the most susceptible organism tested, with *B. globiforme* and *A. scabies* following in decreasing order. This finding may be related to the fact that both *C. sepedonicum* and *A. scabies* are Gram-positive, while *B. globiforme*, in the characteristic coccoid stage, which it rapidly assumes on transfer, is also predominantly Gram-positive. It is not to be inferred that the degree of inhibition follows the same order as that shown in Fig. 1, for from Table I we may observe that *R. japonicum* when antagonized showed relatively large zones, yet this organism was acted upon by the least number of actinomycetes.

A further point of interest brought out by the survey is the relationship of the intensity of antibiotic effect exerted on the test organism to the numbers of test organisms acted upon by the actinomycetes. From Fig. 2 it is seen that those actinomycetes having the most intense antibiotic activity are, in general, the most versatile and act upon the greatest number of bacterial species.

The use of a single culture medium for the demonstration of antagonism is not without limitations, for antibiotic production is known to be regulated by many factors in the nutriment supplied. Since the antagonisms were

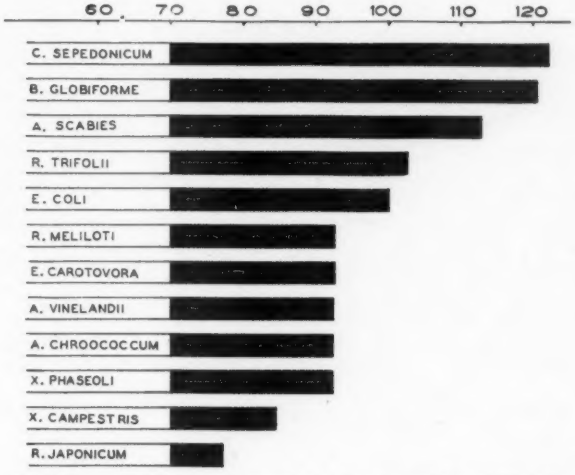

FIG. 1. *Relative susceptibility of test organisms to antagonistic actinomycetes (E. coli = 100).*

evident on a simple synthetic medium even greater effects might be expected under more favorable and varied conditions. Results based on a study of

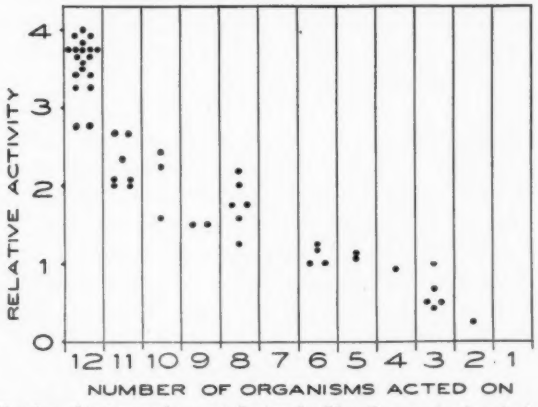

FIG. 2. *Number of test organisms acted upon by 50 actinomycetes in relation to intensity of antibiotic effect.*

the relationship of two isolated cultures are not necessarily applicable to soil with its heterogeneous microflora and vast possibilities of greater interactions. Though organisms possessing antagonist properties against a wide variety

of species may prove important in the establishment of bacterial balances in soil, yet those with greatest specificity may play an important role in the possible suppression of definite types, such as plant pathogenic soil-borne organisms.

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GROWTH STUDIES OF NORMAL AND PARTHENOCARPIC TOMATO FRUITS[1]

By K. A. Clendenning[2]

Abstract

Gaseous exchange by growing tomato fruits is localized in a ring of lenticel tissue that encircles the receptacle and is covered by the calyx. The size of this annular lenticel varies with the size of the fruit.

Growth of the fruits of the variety Grand Rapids includes a phase of residual mitotic activity that persists for approximately one week after setting. Cell division is reduced in dwarf or laggard fruits, which are examples of natural parthenocarpy. Under seasonal conditions that are favorable for natural setting, application of 0.2% indolebutyric – 50 p.p.m. β-naphthoxyacetic acid to pollinated flowers markedly increases the number and size of fruits obtained with the Marglobe but not with the Grand Rapids variety. The locular pulp of parthenocarpic fruits usually remains green during vine ripening. The green color is largely imparted by chloroplasts that are concentrated in the vicinity of the vascular strands leading to the aborted ovules.

Introduction

As a result of numerous investigations, it is now well established that the exposed skin of tomato fruits is devoid of lenticels and stomata at all stages of development (7, pp. 560, 567). The available evidence also indicates that gaseous exchange by mature fruits in storage occurs chiefly in the stem scar region: blocking the stem scar area with wax results in a drastic inhibition of respiration and ripening (2, 3); a slight inhibition of ripening is also effected by leaving the stems attached to the fruit (2, 16). Less is known concerning the region of gaseous exchange in tomato fruits as they grow and ripen on the plant. Measurements of carbon dioxide emission *in situ* (by the mature fruit, calyx, and pedicel up to the abscission layer) revealed a pronounced respiratory climacteric (4). When a ring of wax was applied around the base of the stems, attached fruits did not ripen normally (4).

Information on the developmental stage at which cell division ceases in the flesh of the tomato fruit should improve our understanding of its normal and parthenocarpic growth. If mitosis continues for even a few days after anthesis, the effect of growth chemicals on cell division warrants consideration. Information on mitosis in the young fruit also assists in the interpretation of physiological data relating to successive stages of growth. For the stages in which there are no cell divisions, calculations that are made on a unit fruit basis refer to a constant cell population, whereas data expressed on a weight basis refer to widely different numbers of cells.

[1] *Manuscript received September 8, 1948.*

Contribution from the Division of Applied Biology, National Research Laboratories, Ottawa, and the Department of Horticulture, Ontario Agricultural College, Guelph, Ont. Part of this paper is taken from a thesis submitted to the Graduate School of the University of Toronto in partial fulfilment of the requirements for the degree of Doctor of Philosophy. Issued as N.R.C. No. 1844.

[2] *Biochemist, Plant Science Investigations.*

Agreement has not been reached as to the stage of development at which mitosis actually ceases. Smith (14) reported that cell division proceeds actively in the fruit flesh for approximately two weeks after pollination. Houghtaling (8) as well as Gustafson and Houghtaling (6) concluded that growth after pollination was caused entirely by cell enlargement. MacArthur and Butler (10) reported that ovary growth was entirely by cell division prior to pollination, and that subsequent growth was chiefly by cell expansion, cell division being a minor factor that just sufficed to maintain the epidermis, the cells of which do not expand. Groth (5) had previously reported that young and mature fruits contain the same number of epidermal cells, and that mitosis plays little part in the enlargement of the tomato fruit skin.

The yield and quality of fruits that are obtained following growth chemical applications to intact tomato flowers has been investigated extensively, notably by Howlett and Marth (9). From these and earlier studies, it has been established that growth and maturation can be accelerated, seed production curtailed, and the yield of marketable fruits increased under seasonal conditions unfavorable for natural setting (12). The large increases in yield reported by Howlett and Marth (9) were obtained with early spring populations of unspecified varieties. The interacting effects of growth chemical applications, varieties, and seasonal or environmental factors apparently have not been investigated.

This paper reports observations on the area of unrestricted gaseous exchange in the growing tomato fruit, and on the persistence of mitosis in the fruit flesh after anthesis and setting. The effects of spraying hand pollinated and emasculated flowers of the Grand Rapids and Marglobe varieties with 0.2% indolebutyric – 50 p.p.m. β-naphthoxyacetic acid solution have also been tested under conditions that were judged favorable for natural setting.

Materials and Methods

The greenhouse populations employed in this investigation bore ripe fruit in July. The plants were propagated in soil essentially as directed by Walford (15). Daily tapping of the wire supports was used to effect self-pollination. The flowers were tagged at 'setting', or on the first day that wilting of the corolla was evident. Growth of individual fruits was followed by caliper measurements of the mean equatorial diameter. The number of cell layers composing the pericarp at successive stages of growth was counted radially at the equator. The observations summarized in Table II were made on stained microtome sections from a minimum of six ovaries or fruits. The counts were taken between vascular bundles at four different positions on each section. The cell counts on artificially induced and naturally parthenocarpic fruits were taken in the same manner on razor sections.

The treated and control fruits used in the waxing experiments occupied adjoining, randomized positions on the trusses, and were of similar size and age (diameter 30 to 45 mm., age 18 to 24 days). The sepals were removed

with forceps before the waxes were applied. Calyx removal was found experimentally to have no noticeable effect on subsequent growth and ripening. Hot paraffin (175° F.), and unheated vaseline and lanolin were applied as $\frac{1}{4}$-in. rings around the base of the pedicel.

The Grand Rapids and Marglobe plants (24 of each), employed in the growth chemical experiment, were grown simultaneously in adjoining benches. The treated and control flowers occupied adjoining, randomized positions on the third and fourth trusses. The growth chemical solution was essentially that which was found most effective by Howlett and Marth (9), namely 0.2% indolebutyric acid – 50 p.p.m. β-naphthoxyacetic acid. Carbowax 1500 (5%) was employed as a carrier. The individual flowers were sprayed once, with a nasal atomizer, and were shielded to prevent contamination of the adjacent controls. Maximum self-pollination of the 200 flowers referred to in Table III was effected by shaking and dusting them individually on the day that pollen was being freely shed. This treatment preceded spraying of the exposed flower parts. Flowers also were emasculated and sprayed when half-opened, or approximately two days before pollination would have begun. The growth chemical solution in this instance was sprayed directly on the pistil.

Results and Discussion

Effects of Waxing the Region of Calyx Insertion

All of the wax treatments tended to reduce subsequent growth and to inhibit ripening on the plant (Table I). Growth was stopped completely in two fruits treated with vaseline and in three of the fruits receiving paraffin, but

TABLE I

GROWTH OF TOMATO FRUITS FOLLOWING APPLICATIONS OF WAX TO THE CALYX REGION

(All fruits were waxed 21 ± 3 days after setting at an initial diameter of 30 to 45 mm.)

Treatment	Number of fruits	Average increase in diameter, mm.	
		After two weeks	After four weeks
Control	24	11.9	16.7
Vaseline	12	7.7	9.7
Paraffin	12	6.0	8.5
Lanolin	12	3.9	6.8

complete blocking of subsequent growth was not observed with lanolin. Fruits that showed no subsequent growth assumed a leaden color within one week and finally shrivelled.

The most striking accompaniment of waxing was the appearance of large bubbles, usually within an hour or two, which on bursting provided a large breathing pore through the wax layer (Fig. 1). This phenomenon always occurred when lanolin was used, and the bubbles frequently rose to a height

of ½ to 1 in. above the wax collar before bursting. Repeated plugging of the resulting apertures over a two week period did not prevent fresh holes from appearing within a day or two. Because of this persistent breaking of the seal, the reduction of growth obtained with lanolin must have resulted from partial or intermittent blocking of the region of calyx insertion.

When clear mineral oil was used as the barrier on attached and detached fruits, it was possible to mark the point of bubble emergence with a needle. With detached fruits, bubbles were observed to rise from the ring of vascular bundles within the stem scar zone. On attached fruits, the bubbles always appeared in a ring of uncuticularized tissue that surrounds the receptacle and ordinarily is covered by the calyx (Fig. 2). This tissue forms a massive annular lenticel. Since gas bubbles did not emerge from the sepal scars at the base of the pedicel, the calyx apparently does not function in the respiratory exchange of growing fruits. Longitudinal sections through the lenticel tissue at the point of bubble emergence revealed vascular strands in close proximity. These strands lead from the outer fruit wall to the pedicel, and pass just beneath the annular lenticel.

Examination of mature fruits from different varieties revealed that lenticel ring width varies directly with fruit size. This relationship to fruit size was also evident in Grand Rapids fruits examined with an eyepiece micrometer at successive stages of growth:

Fruit diameter, mm.	10	20	25	35	50	65
Lenticel ring width, mm.	Visible	0.1	0.25	0.5	1.3	1.7

Cell Division in the Pericarp After Setting

Table II provides a summary of our observations on the number of cell layers comprising the pericarp of Grand Rapids ovaries and fruits at successive stages of growth. Cell division continued in the fruit wall after pollination and setting but ceased within the first week. No increase in cell number was

TABLE II

NUMBER OF CELL LAYERS IN THE PERICARP OF GROWING TOMATO FRUITS

Stage of development	Ovary diameter, mm.	Pericarp cell layers
Flowers opening (1 day before pollination)	1.8	9
Flowers 'setting' (2 days after pollination)	3.0	14
After setting		
2 days	6.0	18 – 20
8 to 10 days	20.0	22 – 24
15 to 20 days	35.0	22 – 24
30 to 35 days	50.0	22 – 24

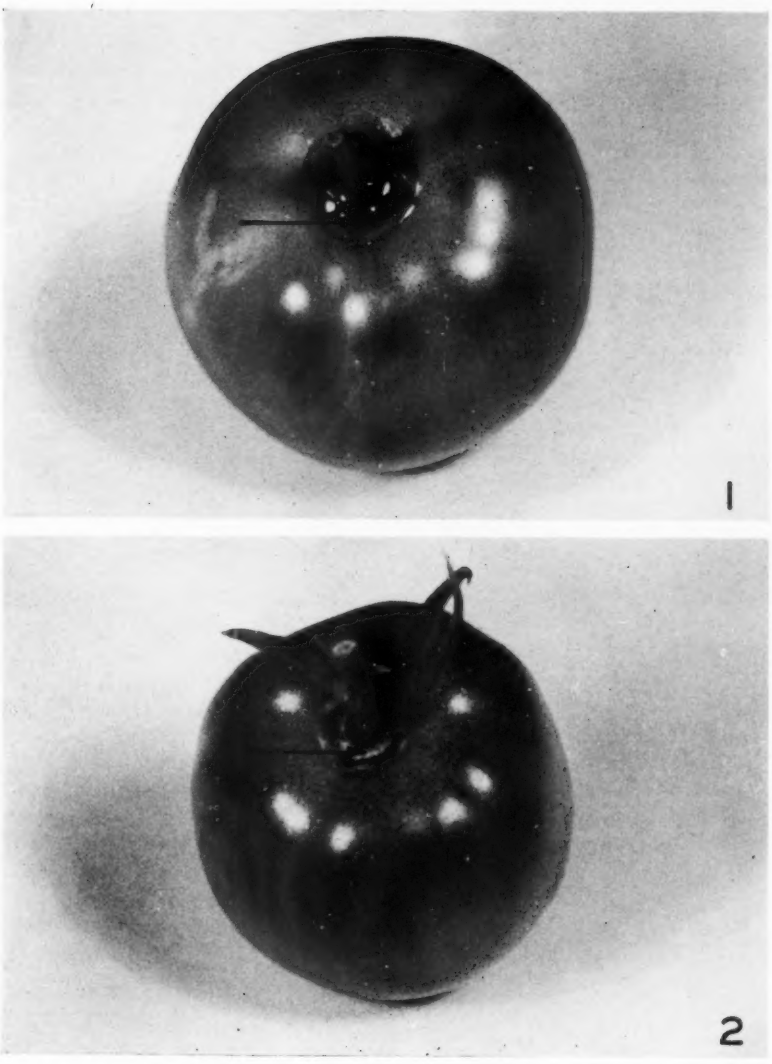

FIG. 1. *Waxed Marglobe fruit, showing a typical breathing pore (arrow) over the annular lenticel.*

FIG. 2. *Unwaxed Marglobe fruit, showing white sepal scar zone at the base of the pedicel (arrow), surrounded by darker lenticel tissue.*

observed in the outer and interlocular walls after the fruits attained a diameter of approximately 1 cm. Moreover mitotic figures were not observed in sections of the placenta and pericarp from two- to three-weeks-old fruits.*

These observations are in general agreement with those of Smith (14), which were made on the Bonny Best variety. His data differ from those of Table II with respect to the final depth of cells composing the pericarp. This is not believed to be a real varietal difference for the following reasons: the camera lucida drawings by MacGillivray (11, Fig. 6) of Bonny Best fruit tissue show the same depth of cells in the mature fruit wall as are recorded above for the Grand Rapids variety; examination of Smith's drawings (14) reveals that his cell counts were taken through the vascular bundles rather than between them as in the present study.

Houghtaling's conclusion (8) was based on the direct proportionality that was found to be maintained between cell volume and fruit volume throughout the grand period of growth. Her data (Fig. 2, p. 249) show, however, that this simple relationship of cell size to fruit size ($k = 1$) does not appear until approximately one week after setting. It therefore appears that among all varieties of *Lycopersicon esculentum* that have been investigated, cell division normally persists in the ovary wall for several days after pollination, and that subsequent growth is entirely by cellular enlargement.

Laggard Fruits

Tomato plants often bear a few fruits that grow very slowly, reach a final diameter of 15 to 30 mm., and ripen at an age of about 60 days. Our observations on approximately one hundred of these fruits (Grand Rapids and Marglobe) indicate that they are always seedless, and that their locular pulp does not remain green during ripening as in the larger parthenocarpic fruits obtained with growth chemicals (*vide infra*). When fully ripened, these stunted fruits have a dull appearance, lacking the luster of normal fruits. Undersized parthenocarpic fruits of the Grand Rapids variety, obtained by smearing ovaries with 0.5 to 1.0% indolebutyric acid in lanolin, had the same external appearance when fully ripened. The number of cell layers in the pericarps of fruits from the naturally seedless class was found to be the same at maturity (14 to 16 layers) as exists in ovaries at the 'setting' stage. The factors responsible for their seedless habit and reduced size thus affect cell division, possibly after setting.

Effects of Growth Chemicals on Fruit Yield and Quality

The experiment reported in Table III was designed to test the effects of spraying pollinated flowers, in late spring, with a growth chemical solution that had previously proved highly satisfactory in early spring applications when natural setting was very poor (9). The two varieties used in the present work differed in their natural ability to set fruits. Under favorable conditions,

* *Truscott, J. H. L. Unpublished observations.*

TABLE III

NUMBER AND WEIGHT OF FRUITS OBTAINED FROM HORMONE SPRAYED AND UNTREATED TOMATO FLOWERS

Fifty flowers per treatment

Treatment	Number of fruits set	Average wt. of fruits, gm.	Total weight of fruits, gm.
Grand Rapids variety			
Pollinated (control)	30	58.5	1756
Pollinated and sprayed	33	55.5	1833
Emasculated and sprayed	9	44.1	397
Marglobe variety			
Pollinated (control)	9	89.7	807.5
Pollinated and sprayed	25	147	3675
Emasculated and sprayed	20	110	2196

the Grand Rapids variety sets fruits abundantly. Although Marglobe is one of the most important canning and shipping varieties grown in the United States (13), its unreliability with respect to fruit setting is a serious shortcoming (1).

All fruits from the control Grand Rapids flowers contained large numbers of seeds in each locule. Spraying pollinated Grand Rapids flowers reduced the number of fruits containing seeds, but increased the total number and total weight of fruits obtained. The average weight of individual parthenocarpic fruits obtained from either pollinated or emasculated Grand Rapids flowers was less than that of the fruits containing seeds. As a result of this spray treatment, few of the emasculated Grand Rapids flowers bore fruit of marketable size.

The Marglobe variety was characterized by lower natural setting, and higher artificial setting of emasculated and pollinated flowers than was observed in the adjoining Grand Rapids population (Table III). The final size was consistently greater for the parthenocarpic Marglobe fruits obtained from emasculated flowers than for Marglobe fruits containing seeds. The majority of the fruits obtained from Marglobe flowers that had been both pollinated and sprayed were without seeds, and the remainder contained approximately one seed per locule. The yield of fruits from pollinated Marglobe flowers was increased approximately four times by spraying, despite the lateness of the season and restriction of the treatment to hand-pollinated flowers of middle trusses. With this variety at least, the yield may be improved by the use of growth chemicals, even when the light conditions are favorable for the production of viable pollen.

The parthenocarpic fruits were indistinguishable in external appearance from the fruits containing seeds and neither class showed appreciable 'puffiness'. The seedless and normal fruits showed equal numbers of cell layers in the pericarp, which indicates that mitosis after setting was not affected by the growth chemicals. The parthenocarpic fruits of both varieties were characterized by the retention of green color in the locular pulp during vine ripening. The ripening color change in fruits containing seeds usually begins in the locular pulp before it is shown by the exposed parts. The parthenocarpic condition seems to reverse the ripening sequence, i.e. from the outside in, rather than from the inside out. Microscopic examination of the locular pulp of ripened and partially ripened parthenocarpic fruits revealed that the green color is largely imparted by intact chloroplasts. These are particularly numerous around the free ends of the vascular strands that normally serve the seeds.

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THE MINERAL REQUIREMENTS FOR FLUORESCIN PRODUCTION[1]

By Joyce V. King,[2] Jack J. R. Campbell,[3] and Blythe A. Eagles[4]

Abstract

An investigation of the salt requirements for fluorescin production has been carried out, and the mineral balance for maximum fluorescin production determined. Employing ammonium succinate as a source of nitrogen, the ions PO_4, Mg, SO_4, and Fe have been found essential to the formation of the pigment. Optimum production of fluorescin resulted when 0.01% magnesium chloride, 0.06% dipotassium hydrogen phosphate, 0.005% potassium sulphate, and 0.000005% (0.05 p.p.m.) ferric chloride were used.

The influence of high concentrations of Mg and SO_4 ions in the presence of varying concentrations of iron with ammonium succinate as the source of nitrogen on the formation of both fluorescin and pyocyanin has been determined.

Fluorescin production has been demonstrated to depend primarily upon the concentration of sulphate, iron, and magnesium in the medium. The concentration of iron required has been shown to be of paramount importance. Increasing the amounts of either SO_4 or Mg did not bring about the pronounced stimulation of fluorescin production as was evidenced in the case of pyocyanin.

Introduction

In studies on the amino acid and mineral requirements for pyocyanin production by *Pseudomonas aeruginosa*, the required salt balances for maximum production of pyocyanin employing specific amino acids as a source of nitrogen have been established (2 and 3). Although certain conclusions could be drawn with respect to the conditions governing the production of bacterial fluorescin no attempt was made to investigate thoroughly the factors relating to the formation of this green fluorescent pigment.

Various media for the production of the pigment have been proposed. In his work on pyocyanin formation Aubel (1) noted that a fluorescent pigment was produced by *Pseudomonas pyocyaneus* but thought its formation to be a caprice of the organism. However, Sullivan (6) contended that an alkaline reaction and high phosphate content favored fluorescent pigment production while low phosphate and an acid reaction favored pyocyanin formation. In 1932 Robinson (5) described a synthetic medium for fluorescin production and in 1942 Jamieson (4) showed that the ions SO_4, Mg, and PO_4 are essential to its production.

The object of the present investigation was to establish the conditions optimum for fluorescin production in the absence of pyocyanin formation and in the light of the data thus obtained to clarify the behavior of the organism with respect to the formation of both pigments.

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Methods

The organism employed in this work was *Pseudomonas aeruginosa* A.T.C. 9027.

In order to determine the influence of the respective constituents of the medium on fluorescin production a series of flasks were prepared, the contents of which differed only by the concentration of the variable under test. For the preparation of media concentrated stock solutions of the various constituents were prepared and preserved under toluene. Basal media of twice the final concentration were prepared, the variable constituent was added, and the medium was then made to volume with water. All media were made up in 10 ml. quantities in 125-ml. Erlenmeyer flasks. Reagent chemicals were used and all solutions were made up with water redistilled through glass. The 125-ml. Erlenmeyer flasks were covered with 100-ml. beakers. Glassware was thoroughly treated with aqua regia and rinsed with water redistilled through glass. Media were sterilized at 15 lb. pressure for 15 min.

The inoculum was prepared by transference of a loopful of a 24 to 48 hr. culture of the test organism into 10 ml. of Sullivan's medium and incubation at 30° C. until the appearance of faint turbidity—usually about 24 hr. The cells were then centrifuged and resuspended in 10 ml. of sterile physiological saline.

The extent of fluorescin production was determined by means of a fluorimeter employing a 1 : 25 dilution of the chloroform insoluble fraction of the medium in calibrated tubes. Dichlorofluorescin was used as standard. Pyocyanin was measured as previously described (3).

Experimental

In studies on the effect of the nitrogen source on fluorescin production the basal medium consisted of 0.1% dipotassium hydrogen phosphate, 0.1% magnesium sulphate septahydrate, 0.05 p.p.m. ferric chloride, and 1.0% glycerol.

In studies on fluorescin production uncomplicated by the elaboration of pyocyanin it is essential that the amount of iron added to the medium be kept at a low level, approximately 0.017 p.p.m. (0.05 p.p.m. ferric chloride); the amount of iron present in the medium from other sources is estimated to be 0.009 p.p.m.

When using, as the source of nitrogen, potassium nitrate, sodium nitrate, or ammonium sulphate at a concentration of 0.02 moles of nitrogen per liter, fair growth was obtained but there was no evidence of pigment formation even after four days' incubation in these media.

The ammonium salts of various dicarboxylic acids were employed as sources of nitrogen—oxalic, malonic, succinic, glutaric, adipic at a concentration of 0.025 moles of nitrogen per liter, respectively. In this series the dipotassium

hydrogen phosphate and magnesium sulphate were employed at 0.05% concentration. Growth was obtained in all media but in the case of ammonium oxalate no pigment was formed.

When the determinations were carried out after two days' incubation the pigment formation from ammonium succinate was greater than that observed in the other sources of nitrogen; after four days no difference in the amount of pigment formed in the various media was observed.

Ammonium succinate was shown to be the most suitable source of nitrogen for the development of fluorescin, and higher concentrations were shown to be without significant influence on pigment production. A gradual lessening in the amount of pigment formed proportional to the variation in chemical structure from ammonium succinate was observed.

For the determination of the comparative effect of ions essential to fluorescin production media containing varying concentrations of dipotassium hydrogen phosphate, magnesium chloride, and potassium sulphate were prepared. The salt concentrations employed and the amounts of fluorescin and pyocyanin obtained from the respective media are recorded in Table I.

TABLE I

THE EFFECT OF SALT CONCENTRATION ON FLUORESCIN AND PYOCYANIN PRODUCTION

K_2HPO_4, %	$MgCl_2$, %	K_2SO_4, %	Fluorescin*	Pyocyanin**
0.05	0.01	0.0004	0	0
0.05	0.01	0.0005	Trace	0
0.05	0.01	0.002	18	0
0.05	0.01	0.008	18	0
0.05	0.01	0.01	18	0
0.05	0.01	0.1	17	0
0.05	0.01	0.2	14	0
0.004	0.01	0.005	12	3.5
0.008	0.01	0.005	13	4
0.03	0.01	0.005	18	0
0.08	0.01	0.005	19	0
0.1	0.01	0.005	17	0
0.05	0.001	0.005	0	0
0.05	0.004	0.005	14	0
0.05	0.01	0.005	18	0
0.05	0.05	0.005	12	0
0.05	1.00	0.005	0	0

In Table I the amount of fluorescin formed is shown to depend on the concentration of dipotassium hydrogen phosphate. At lower concentrations of phosphate with resulting diminished conditions of growth the low concentration of iron in the medium appears to be released for the formation of small quantities of pyocyanin.

* *Fluorescin expressed in terms of percentage of dichlorofluorescin* $\times 10^4$.

** *Pyocyanin expressed as reading on logarithmic scale of Fischer Electrophotometer.*

Magnesium is shown to be essential for fluorescin production, which is seen to depend upon the concentration of this ion in the medium, Table I. As will be shown later, Table III, the influence of magnesium on fluorescin production is in a measure determined by the relative concentration of the sulphate ion.

Although the sulphate ion is not required for growth its presence in the medium is essential for fluorescin production. The optimum concentration would appear to lie within the range of 0.002 to 0.01% potassium sulphate.

In order to determine the optimum concentrations of the respective salts required for fluorescin production, media containing these salts at varying concentrations were prepared. The results obtained are recorded in Table II.

TABLE II

OPTIMUM SALT CONCENTRATIONS FOR FLUORESCIN PRODUCTION

K_2HPO_4, %	$MgCl_2$, %	K_2SO_4, %	Fluorescin	Pyocyanin
0.03	0.01	0.005	18	0
0.05	0.01	0.005	19	0
0.06	0.01	0.005	20	0
0.08	0.01	0.005	19	0
0.03	0.16	0.005	17	0
0.03	0.010	0.005	18	0
0.03	0.008	0.005	17	0
0.03	0.005	0.005	16	0
0.03	0.01	0.003	19	0
0.03	0.01	0.005	19	0
0.03	0.01	0.008	17	0
0.03	0.01	0.01	15	0

The production of fluorescin is seen to be maintained at a high level over a wide range of concentrations of phosphate, magnesium, and sulphate ions. On the basis of these findings, a medium most favorable to fluorescin production was evolved as follows:

Ammonium succinate.....0.03%
Glycerol.....1.0%
Ferric chloride.....0.05 p.p.m.
Magnesium chloride.....0.01%
Dipotassium hydrogen phosphate.....0.06%
Potassium sulphate.....0.005%

In the work of Burton *et al.* (2) it was shown that either the magnesium or sulphate ion may in a measure substitute for one another in a medium designed for maximum pyocyanin production in which amino acids serve as the source of nitrogen. In the light of the findings reported herein in which ammonium

succinate was successfully employed as a source of nitrogen, it was considered desirable to determine whether or not results comparable with those reported by Burton *et al.* would be obtained. With this end in view the experiments detailed in Table III were carried out under conditions favorable to pyocyanin formation without fluorescin production. Parallel experiments on fluorescin formation in the absence of pyocyanin production were also performed. The results are recorded in Table III.

TABLE III

THE EFFECT OF HIGH CONCENTRATIONS OF MAGNESIUM AND SULPHATE IONS ON PIGMENT FORMATION

$MgCl_2$, %	K_2SO_4, %	(High iron—10.0 p.p.m. $FeCl_3$)		(Low iron—0.05 p.p.m. $FeCl_3$)	
		Pyocyanin	Fluorescin	Pyocyanin	Fluorescin
0.5	0.005	35	0	0	15
0.5	0.18	79	0	0	13
0.5	0.6	94	0	0	13
0.1	1.0	57	0	0	18
0.3	1.0	100	0	0	17
1.0	1.0	85	0	0	0

The findings of Burton *et al.* with respect to pyocyanin are confirmed. In the case of fluorescin, increasing the concentrations of sulphate ion did not exhibit the pronounced effect on pigment production observed for pyocyanin. With magnesium, similar results were obtained within certain concentrations of the ion. Above 1% magnesium chloride complete absence of fluorescin production was observed even though marked growth occurred.

In the light of the findings of Burton *et al.* on the interrelationships of magnesium, sulphate, and iron on pyocyanin formation and with the object of lessening the possible influence of trace contaminants present in the high concentrations of salts employed by these workers, experiments were carried out using lower concentrations of these ions. In the presence of an adequate supply of iron, pyocyanin would be produced and in view of the results recorded in Table II in which 0.01% magnesium chloride, 0.005% dipotassium phosphate, and 0.05 p.p.m. ferric chloride provided a good medium for growth and gave high yields of fluorescin, experiments were carried out employing similar concentrations of magnesium and sulphate in the presence of higher quantities of iron. The results of these experiments are recorded in Table IV.

In Table IV, the results obtained when the concentrations of magnesium and sulphate were varied over a considerable range with the concentration of iron held at a uniformly high level of 0.001% (10 p.p.m.) ferric chloride are detailed.

Good growth was obtained in all media. Fluorescin was absent under all conditions thus confirming previous observations. The results show clearly that pyocyanin formation depends primarily upon the quantities of sulphate

and iron present in the medium and that although magnesium is required for pyocyanin formation its essential function is concerned with the growth of the microorganism.

TABLE IV

THE ROLE OF MAGNESIUM, SULPHATE, AND IRON IN PYOCYANIN PRODUCTION

$MgCl_2$, %	K_2SO_4, %	$FeCl_3$, %	Pyocyanin	Fluorescin
0.01	0.005	0.001	0	0
0.01	0.03	0.001	18	0
0.01	0.05	0.001	33	0
0.02	0.005	0.001	0	0
0.1	0.005	0.001	0	0
0.3	0.005	0.001	0	0

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SEEDS AND SEEDLINGS OF THE GENUS *BRASSICA*[1]

By Jean M. McGugan[2]

Abstract

The genus *Brassica* with its numerous cultivated strains and weed varieties presents a difficult problem to the seed analyst. A solution is attempted in this paper. Peculiarities of seed color, shape, and histology have been used wherever possible as diagnostic characters. In cases where these were found to require verification the characters of the young seedlings were used to supplement them. It has been found possible to distinguish the seeds or seedlings of plants so closely related as to be separated in adult form only by differences in the depth of the yellow color in their roots and blossoms. Care has been taken to test the material for authenticity and to make sure that the seed and seedling characters chosen are closely linked in heredity with distinguishing characters of the species and varieties. A key for the identification of seeds of known varieties of *Brassica* has been evolved.

I. Introduction

The term "Brassica" was used by the Greeks as early as 200 B.C., and was first applied to the cabbage group of plants, *Brassica oleracea* L., together with the radish, *Raphanus*—plants which indeed had already been in use for several hundred years. It is thus noted in Cato's "De Re Rustica", 149 B.C., but, since then, its scope has gradually broadened and it has now become a genus of widespread economic importance, ranging from the much desired foodstuffs of animal and man to some exceedingly undesirable and more or less poisonous weeds, and is probably the most outstanding genus of the Cruciferae.

Linnaeus, in his eighteenth century classification, distinguished from the genus *Brassica* a genus *Sinapis*, which was originally intended to represent the true mustards. It was found, however, that mustard principles were prevalent throughout, and as they varied in concentration from that sufficient to add piquancy to salad greens to that adequate to supply condiments, counterirritants, and rubefacients for commercial use, they formed only a relative basis of classification. During the nineteenth century there was a concerted movement to eliminate the genus *Sinapis* and include all of its members in the genus *Brassica*. This later classification is commonly followed in Canada. Engler's "Das Pflanzenreich", however, in the section on Cruciferae–Brassiceae compiled by O. E. Schulz in 1919, still retains the Linnaean classification but bases the distinction on the lateral nerves of the fruit. In his work 35 species are placed in the genus *Brassica*, while *Sinapis* comprises seven of which two are *alba* and *arvensis*. It might be mentioned here that Kamenskei

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(1931 (28)) claims that in the seed coat of the genus *Brassica* the large cells and the epidermis compress into one layer while in *Sinapis* the epidermis is distinct. The writer has not found that this distinction is valid in all cases and in the present work all species are taken as belonging to the one genus, *Brassica*.

As might be expected, a genus as large as this one is represented by a variety of forms, but the genus *Brassica* exhibits some unusual modifications. Worthy of special note is the fact that either the flower, the shoot, the stem, the root, the leaves or parts or combinations of these may develop characteristically while the remaining vegetative organs of the plant remain normal. This development is marked in *B. oleracea* L. In the variety *botrytis* L., the whole flower structure, but principally the peduncle, enlarges to form the cauliflower or, in a later strain, the broccoli head. In the variety *capitata* L., the stem becomes so dwarfed that the leaves fold over each other in a solid cabbage head. In the variety *gemmifera* DC., Brussels sprouts, individual shoots form themselves into small heads in the axils of the stem leaves. In the variety *caulorapa* DC., kohlrabi, the stem becomes swollen, and in the variety *acephala* DC., the kales, the leaves and the stem may become slightly swollen, or the leaves become variously cut and curled. Similar tendencies are seen in other species. In *B. pekinensis* Rupr., pe-tsai, the petioles and veins of the leaf become variously modified. In *B. napobrassica* Mill., rutabaga or Swede turnip, the lower part of the stem and the root form themselves into an enlarged schizocorm, and in *B. Rapa* L., the turnip, the root alone becomes enlarged. In many forms, however, modifications are not apparent or, if present, are only relative and not distinct.

Cause for these modifications may be found in the following contributary factors: (1) within the genus variations frequently occur and polymorphy is common, (2) improvement of type through selection is constantly being maintained, and (3) species crosses, as well as intergeneric crosses, are possible, and many variety crosses occur, as Kristofferson (1924 and 1927 (33, 34)), Kakizaki (1925 (27)), Karpetchenko (1924 and 1927 (29, 30)), Sinskaja (1927 (58)), and Malinowski (1929 (39)) have shown. Independent or multiple hereditary factors may be involved. Red color of cabbage, for example, as well as certain other color units, behaves as a simple Mendelian character, whereas the hearting of cabbage, the curling of the kale leaf, and the shaping of the storage organs are the result of multiple factor groups. The complication of the picture may be increased by complete or partial linkages, as well as multiple linkages where the individuals from two groups of multiple factors are themselves linked.

On the other hand two species that have followed different lines of development may contain certain identical factors and in consequence show surprising similarities. Note the striking resemblance between the enlarged storage

organ of certain strains of rutabaga and turnip (Kajanus, 1917 (26)). Considering that these two plants belong to different species, have different colored flowers, and a different number of chromosomes (*B. Napus* $2n = 38$ and *B. Rapa* $2n = 20$, Howard, 1940 (24)), this likeness might easily cause confusion. Conversely allelomorphic factors are also to be found in these two species, the hairiness of the rutabaga leaf and the smoothness of that of the turnip (Malinowski, 1929 (39)).

Considering that it is on distinctions of the above order that plant classification is necessarily based, one need feel no surprise that different systematists have differed so radically in their *Brassica* groupings; the resultant confusing nomenclature of the genus* is, however, unfortunate.

In the light of these facts consider the possibility of identifying *Brassica* seeds without destroying them, i.e. by the outer seed coat marking, or the color, size, and shape of the seed itself. Such an identification presupposes that, in seed of each variety, one at least of the above characters is controlled by simple factors completely linked with some factor or factors responsible for the particular identification features of that variety of plant. Undoubtedly many such incidents exist but in certain cases they are lacking.

Another difficulty presents itself to one attempting the identification of *Brassica* seeds by inspection alone. Whether due to multiple factor groups or to some other cause the seeds of one variety quite commonly show a gradation of certain characters that may extend into one or two closely related varieties. This is so general that *Brassica* seeds in common use seem to form themselves into a practically unbroken series from *B. oleracea* through *B. Napus*, *B. napobrassica*, *B. Rapa*, and *B. campestris* to *B. juncea*. Of course there are exceptions to and minor offshoots from this series but if enough varieties and strains are on hand it is surprising how gradual the transition appears. Granting that many of these varieties are clear-cut and distinct and will always remain true to type, there are others which, even when their seeds in bulk lot can be placed in one variety or another, will contain some individual seeds whose diagnosis is by no means clear.

Turning to the literature on the subject, we find that the first studies of the seeds of the genus *Brassica* were undertaken by pharmacologists and at first centered on the hilum and micropyle. Later, and until the end of the nineteenth century, the main efforts were directed toward discovering the layers that make up the seed coat. In the twentieth century work turned to the distinction of *Brassica* seeds by their very young seedlings, and thus naturally pigments were brought into the limelight. This was followed by a study of older cotyledons, and later, of quite well developed plants, with attention being directed to the character and shape of cotyledons and first

* *For a most interesting account of the derivation of Brassica names I would make reference to: "Flora of Middle Europe", by Gustave Hegi, 1906.*

leaves. Some work was done with the use of ultraviolet light. In this literature there is a confusion of scientific names coupled with common names in unfamiliar languages. It thus happens that it is often impossible to take full advantage of the work that has been done, because unfamiliarity with the common varieties of the countries concerned makes it difficult definitely to connect their variety name with ours.

This paper which treats on different phases of *Brassica* identification in turn, is the result of investigation extending over a number of years, during the course of which it has been found that generally speaking when *Brassica* plants themselves are distinctive their seeds may be conclusively identified. On the other hand where the distinction between plant groups is based merely on relative modifications, a corresponding correlation exists with reference to their seeds, which can be distinguished only by a complete study of the seed and seedling from all angles and a careful evaluation of the results.

For such an investigation a broad selection of authentic samples of seed is necessary and in the present work more than seven hundred such samples were used. Of these 85 came from the Central Experimental Farm, Ottawa, where their identity had been certified. Through the courtesy of W. H. Wright, Chief of Analytical Services, Ottawa, I was enabled to study five samples of rape seed identified by Dr. F. H. Hillman, former Assistant Botanist, Seed Laboratories, Washington. The rest of the seeds were obtained from various seed companies, botanical gardens, and government stations at home and abroad. From the majority of these samples plants were raised to maturity and identified as to variety. In this connection my grateful thanks are due to Mr. A. Hope of the Canadian Seed Laboratories at Sackville, N.B., as well as to the Forage Crop Division, Ottawa, for the growing and forwarding of plants of 90 of them. The remainder were grown by the writer who is indebted for aid in their identification to Dr. L. H. Bailey, Cornell University, who very kindly allowed her to study the plants in his herbarium.

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II. Seed Coats

(Their Importance in Seed Identification)

A very thin radial section of the *Brassica* seed coat can be used to identify species and very often varieties. Typical *Brassica* seeds are fairly easily placed but those that are off type require careful study. It should first be

noted that the macroscopic appearance of the seed coat is valuable in the elimination of certain varieties. The thin coat of Brussels sprouts, for example, contrasts definitely with the thick heavy one of marrow kale. Secondly, portions of the seed coat, cleared in chloral hydrate or weak potassium hydroxide, often show many of the same characters as the radial section and are much more easily made available for study. This is particularly useful in observing palisade cells.

In the study of the seed coat the epidermal layers were first noted. Oudemans (1854 (45)) seems to have been the first to interpret the tumbler cell of the palisade layer and he described also a subepidermal layer. Fluckiger (1867 (15, p. 687)) was confused about the palisade and pigment layers while Berg (1865 (3, p. 92)) had only three layers to the coat and these were not clear. Schröder (1871 (55)) embedded seeds in wax for cutting and mounted the sections in glycerine. Tietscher (1872 (61, p. 72)) had several illustrations and noted a layer with intercellular spaces below the epidermis. Fluckiger and Hanbury (1874 (16, p. 141)) used turpentine for mounting and showed closed palisade cells and a pigment layer with firm, solid-walled cells. Sempelowski (1874 (56)), like Schröder (1871 (55)), and at a later date Hartz (1885 (21)), studied seed tissues. He could not distinguish *B. Rapa* and *B. Napus*, contradicted the statement that intercellular spaces are present below the epidermis, and has one of the best of the earliest papers. Hohnel (1875 (23)) also used Schröder's method of embedding in wax in a small hole in a cork for cutting, treated his sections with caustic potash, nitric acid, or Schultze's maceration fluid as desired, and stained with chlorzinc iodide. He distinguished *B. Napus* from *B. Rapa* by the ratio of the length and breadth of the palisade cells, which were uniform in height in *B. Napus* but not in *B. Rapa*. He decried the idea that a study of ontogeny was indispensable in the interpretation of the seed coat. Hartz (1885 (21)) claimed that the lumen of the palisade cells was greater than the double wall in *B. Napus* but not in *B. Rapa*. Nobbe (1887 (43)) did not clearly distinguish separate coats and Wittmack, the same year, investigated oil seeds.

Investigation turned to endosperm tissue and D'Arbaumont (1890 (1)) claimed to have found a new coat adjacent to the aleurone layer that was a remnant of the nucellus not changed into aleurone tissue. Others, such as Cauvet (1888 (9)), Brandza (5), Poisson (50), and Maury (1889 (40), and Atterberg (1899 (2)), disagreed as to the number of layers in the seed coats, these ranging, in their opinions, from three to six. Guignard (1893 (19)) found that the so-called air spaces beneath the epidermis were merely large cells. This was the beginning of the "Grossencellen layer" to which so many references are later made. Burchard (1894–1896) made careful illustrations of radial sections of 12 species. His drawings repay careful study. Pieters and Charles (1901 (49)) claimed to have found differences in the coats of

B. oleracea, *Napus*, *nigra*, and *arvensis* but none between Swede turnip and rape. Böhmer (1903 (4)) and Winton (1906 (64)) described individual seeds. Wisselingh (1919 (65)) explained with illustrations the development of the coats from the integuments and nucellus in *Brassica nigra* and *alba*, *Cheiranthus*, and *Cochlearia*. He states that sometimes the inner cuticle, between the inner integument and the nucellus, remains and becomes a "cork layer" dividing the pigment layer from the endosperm. Viehoever, Clavinger, and Ewing (1920 (63)) described Chinese colza found in America. Muravieva (1928 (41)) illustrated head cabbage, Swede turnip, two varieties of turnip, brown mustard, and wild mustard. Her outer parenchyma layer seems to be what this paper calls the outer subepidermal row, and she has drawn all species with transparent intercellular spaces above the lignified cups, having missed the thin walls that limit the palisade cells in this region. Roberts and Thomas (1933 (52)) have outlined a key of radial sections of some Cruciferae. They based their first distinction on the species *alba*, *arvensis*, and *nigra* having epidermal cells "containing a large amount of visible mucilage", and another on the absence of a subepidermal layer in *B. rapa* L. and *B. napobrassica* Mill. With these as distinctions I found it impossible to agree. A paper by Orechova and Lapinskaja (1936 (44)) picturing radial sections of varieties of *B. oleracea* should be noted. They distinguished red cabbage from white by the irregularity of the outer row of cells in the seed coat of the former. The anthocyanins in the seven-day-old seedling would seem to be a simpler means of distinction.

The effect of ultraviolet light on *Brassica* seeds was investigated by Chmelar (1925 (10)) and Gentner (1920 (17)). Ferguson and Ashe (1928 (14)) used polarized light on scrapings from the outer layer of the seed coat. They found that the mucilaginous substances were optically active in varieties of *B. oleracea* L. but not in other forms.

This completes the rather voluminous literature on *Brassica* seed coats in so far as it is known to the writer. It indicates that in the structure of the seed coats we have a means of identification useful in some varieties though not in all. However, the use of various unidentified variety names by authors in different countries and the lack of agreement between workers as to various details of seed coat histology make a complete single investigation of known varieties necessary, if such characters are to be used to full advantage.

Such an investigation has been made, and from it the following conclusions found to be of practical taxonomic use have been drawn.

The radial section of *Brassica* seed coat has the following layers, the distinction of which may be more clearly understood by reference to Fig. 16, where all layers are clearly distinguishable.

1. *Epidermis*

A single layer of very thin cells, five- to seven-sided in surface view.

2. *Subepidermal Layer*

The layer between the epidermis and the palisade layer, the walls of which become blue when tested for cellulose with iodine and sulphuric acid. In some varieties the outer rows seem to have been crushed or partly crushed by pressure of the maturing seed from within, so that the horizontal walls appear as bands tightly pressed together. In others, the outer row is obviously celled. Each of these cells is filled with a mucilaginous material. In radial section this material is homogeneous in *B. arvensis* (L.) Rabenh., but appears to radiate from a cone-shaped structure formed in the center of the base of the cell in *B. oleracea* L. or from a central concentrated threadlike structure stretching from the top to the bottom of the center of the cell in *B. Rapa* L. *rapifera* Metz., shogoin turnip. In *B. alba* (L.) Rabenh., it forms in concentric rings around a central core. The subepidermal layer is not always stretched tightly over the seed. It may be crushed in folds, undulating or otherwise irregular and is, no doubt, responsible for some minor markings on the outside of the seed coat.

3. *Palisade Layer*

A row of cells whose bases and sides to within a greater or less distance from the top have become thickened and lignified so that they appear as heavy tumblers or mugs. This thickening and lignification occurs on the inside of the cell wall. Above the lignified portion this wall, so thin that even the double wall of adjacent cells appear, in radial section, as a sinuous thread, is short and barely discernible in some species, but elongated and individually conspicuous in others. In *B. nigra* (L.) Koch., for example, its arrangement is almost wholly responsible for the primary net that lies on top of the seed coat. Netting caused by variation in height of the lignified portion of the cell wall occurs in other species but it has more the appearance of being a part of the seed coat. In such seed coats the tallest groups of lignified double walls are responsible for the primary net whereas shorter intermediate groups cause the less conspicuous secondary net apparent in the coat of some varieties.

It is interesting to note trends of variation in palisade structure appearing in the genus. *B. oleracea* L., one of the first of the species if taken as listed in Section IV of this paper, has square tumblers with uniform, thin, straight-sided walls of uniform height, Fig. 4. Uneven, heavier lignification of wall becomes apparent in the tumblers of *B. Napus* L., Fig. 6, and of *B. Rapa* L., Fig. 8. In subsequent species the tendency is for groups of these unevenly thickened tumbler walls to become elongated and with smaller bases form tall cylindrical tumblers with comparatively thick walls. These may be found in *B. campestris* L., Fig. 10 and Figs. 12, 14; *B. juncea* (L.) Coss., Fig. 15; and *B. nigra* (L.) Koch, Fig. 17. The palisade layer of *B. arvensis* (L.) Rabenh., at the end of the series, is made up entirely of very tall cylindrical cells of almost equal height and with walls thickened so that there is practically

no lumen. But while there is this general trend of change, each species has its characteristic cell structure, which may be used to good advantage in the identification of the seed.

4. *Pigment Layer*

Often appears as a solid layer of pigment. Histologically it is the inner integument. In approaching that portion of the seed immediately surrounding the hilum this layer gradually becomes wider until, made up of several rows of cells, it is many times the apparently normal width of the layer as noted on the opposite side of the seed. In the full grown seed it is difficult to interpret. Fluckiger (1867 (15, p. 687)), Viehoever, Clavenger, and Ewing (1920 (63)), and Muravieva (1928 (41)) claim that in the plants that they describe this layer is normally one cell thick; Hartz (1885 (21)), Arbaumont (1890 (1)), Pieters and Charles (1901 (49)), Wisselingh (1919 (21)), Winton (1906 (64)), and Douven (1935 (12)) claim it to have one row in some varieties and two or more in others, whereas Hohnel (1875 (23)) and Fedosseyeva (1936 (13)) describe it with more than one row. I would conclude that while pigment color might be noted it is not expedient to use the cell structure of the pigment layer in the distinction of *Brassica* seeds. Fedosseyeva, however, has found that the seeds of "*B. chinensis* L. var. *parachinensis* and *B. juncea* L. var. *sareptana*" have bodies, round from above but oval in cross section, appearing in their pigment layers that are not present in "*B. glauca* Kew". The presence of these bodies in *B. pekinensis* Rupr., chihili, has been observed in my material. As they are not present in *B. Napus* L. they might constitute a distinguishing feature between *B. Napus* L. and *B. pekinensis* Rupr.

The outer aleurone layer of the endosperm has cells that in radial section are more or less rectangular although not always regular. They usually have bulging outer and inner walls. The inner endosperm layers of cells beneath the aleurone layer are crushed and are not figured in this work.

In preparing the radial sections for the present work, the seed was attached by means of paraffin to a wooden block that fitted into a sliding microtome. It was found that the seed was held with sufficient firmness if two-thirds of its thickness projected above the wax. The knife was held obliquely in the microtome and the cutting was done as in the celloidin technique. No water was allowed to reach the seed coat, absolute alcohol being used entirely. The section was transferred to glycerin and mounted unstained in glycerin jelly. In temporary mounts the cellulose was stained blue with iodine and a solution of sulphuric acid (60%) with glycerin (40%).

Following is a summary of distinctive seed coat characters of different species and varieties of the genus *Brassica*. The camera lucida drawings in Figs. 3 to 17 from varieties selected as typical will, it is hoped, serve further to clarify these points. It will be noted that seed coat structure may be used in the distinction of most species but is satisfactory only in the case of a few specific varieties.

Brassica SEED COATS

	Surface view	Subepidermal layer (radial)	Palisade cells (radial)
B. oleracea L. (general)	No pronounced net because of equal height of tumbler walls and of upper limiting wall of palisade cell. Elevations caused by subepidermal layer might be seen in seed coat but not in prepared slide	Outer row generally celled. Cells of different height but each has cone-shaped projection in middle of base from which mucilage contents of cell seem to radiate	Side walls of tumbler cells straight, lignified, and pigmented, uniform in height and thickness; lumen large
1. var. *ramosa* (DC.) Alef. Thousand-headed kale	Seed coat thick	Outer row with cells lower than in Fig. 4	Tumbler cells large and heavily lignified
2. var. *acephala* DC. *a*. Green or variegated kale	Thin seed coat	Outer row with uniform cells	Tumbler cell walls slightly collapsed and undulating in height. Have right-angled flare at top
b. Blue curled Siberian kale *c*. Cottagers kale	Seed coat thicker	Outer row with uniform cells	Similar to green kale but upper half of tumbler is larger than lower and double wall more pointed
d. Marrow kale	Thick seed coat	Thick with shallow-celled outer row	Tumbler side walls of the large cells same height, flat brim and base, not quite perpendicular. Upper wall long. Pigment prominent
e. Asparagus kale		No conspicuous cells in outer row	Tumbler side walls narrower, higher, and straighter than in Fig. 4
f. Ragged Jack kale		No conspicuous cells in outer row	Tumbler cells almost square with side walls even, thin, straight, flaring at top at angle of 120°
g. Sheep kale	Thin seed coat	No conspicuous cells in outer row	Tumbler cells uniformly straight, tall, and narrow. Lumen same width as double wall, which is slightly rounded at top
3. var. *gemmifera* DC. Brussels sprouts Figs. 3, 4	Lumen of tumbler cells large and oval. Epidermis six-sided. Seed coat very thin	Outer row unevenly celled	Tumbler cell walls thin and slightly crushed
4. var. *capitata* L. Cabbage		Outer row has tall cells. In red cabbage these are uneven	Tumblers, with walls thinly lignified and lumen twice the width of double wall, fit tightly together. Upper palisade wall long. Pigment layer thick

Brassica SEED COATS—*Continued*

	Surface view	Subepidermal layer (radial)	Palisade cells (radial)
5. var. *botrytis* L. subvar. *asparagoides* DC. subvar. *cauliflora* (Gars.) DC.	Thin seed coat	Outer row celled	Tumbler walls thicker and straighter-sided than in Brussel sprouts. Lumen and double wall same width
6. var. *gongylodes* L. Kohlrabi	Thick heavy coat	Outer row celled	Tumbler cell walls straight, of same height, with right-angled bases and tops. Lumen narrower than double wall
B. alboglabra Bailey		Thick, even, shallow cells in outer row	Indications of grouping of 2 or 3 elongated cells to form net. Tumbler cells like cauliflower
B. pekinensis Rupr.	Note round bodies in pigment layer. These bodies are oval in radial section	Crushed	Palisade cells like *B. Napus* but taller, straighter tumbler and higher upper wall. Broad lumen wider at top
B. Napus L. Dwarf Essex rape Figs. 5, 6	Fig. 5 is view at top of tumbler cells. Line between the outer and inner limiting cell walls represents the apex of conical thickening. Netting caused by elongated tumbler cells as well as the narrowing of the lumen	Crushed	Tumbler cells broad and angular with a flared and turned back brim. A lignified conical thickening occurs just inside the brim. This along with the one from the next cell forms a crater at their point of junction. Upper palisade wall short. Tumblers slightly higher than broad, have lumen narrower than the double wall
B. Napus L. Annual form	Resembles biennial form	Crushed	Walls taller and thicker with the depression of smaller diameter
B. napobrassica (L.) Mill.	Resembles *B. Napus*	Crushed	Resembles *B. Napus*. Lumen of tumbler varies but same width as double wall
B. Rapa L. Turnip Figs. 7, 8	Focusing down from the brim of the tumbler cell the aperture gradually decreases to constancy. Fig. 5 is view halfway down. The middle line is the curved wall in sharp focus while the outer and inner walls are slightly out of focus, one above and one below	Crushed	Tumbler cell wall thinner than *B. Napus* and thicker than *B. campestris*. The more gradual thickening below the brim gives a rounded appearance to double wall. Upper palisade limiting wall longer than in any closely related variety
Seven-top turnip		Crushed	Tumbler cell walls thinner than turnip, but like it the tumblers press together at the top and separate near to the bottom. Ridges closer and regular

Brassica SEED COATS—*Continued*

	Surface view	Subepidermal layer (radial)	Palisade cells (radial)
Shogoin, tennoje Japanese foliage turnip		Outer row deeply celled each with a central perpendicular axis from which the cell contents radiate horizontally	
Tendergreen, Komatsu-na Mustard spinach		Crushed	Tumbler cells of uneven height with straight walls of uniform medium width, rounded at base. Lumen same width as double wall in the broad cells but narrower in the tall
B. chinensis (L.) Bailey Celery mustard		Crushed, thick	Tumbler cell of uneven height with thick wall increasing in width toward the almost squared top. No depression
B. campestris L., wild form Figs. 11, 12	Lumen of tall cells of the net is very small but of others it is much larger	Outer row not visibly celled. Dips down into lumen of tumbler cell	Tumbler cells thin-walled of uniform height. Upper palisade lateral wall very short. Resembles *arvensis* in the packing of tall palisade cells
B. campestris L., annual form Figs. 9, 10	Each cell has small lumen. Netting is caused by height of cell along with subepidermal layer	Crushed	Primary net sometimes the result of a single double wall. Upper palisade wall very short
B. campestris L., biennial form Figs. 13, 14	The crushing together of a few palisade cells giving appearance of a heavy wall is, along with the height, responsible for netting	Does not dip down into lumen of tumbler cells	Tumbler cells have thin even walls, wide lumen, and are *not* crowded together
B. elongata Ehrh.		Celled outer row	
B. juncea (L.) Coss. General view		Celled outer row	Three or four of thin-walled tumbler cells growing taller and crowding together at top cause the primary net. Cutinized layer between pigment layer and endosperm
a. Wild form Fig. 15		Outer celled row unusually high	Double lateral tumbler wall high and pencil-shaped
b. Curled mustard		Outer celled row medium high	Lumen broad at base, narrow halfway up, and broad at top
c. Smooth leaf mustard Fig. 16		Outer celled row very shallow	Conspicuous for the elongated and wavy upper walls of the palisade cells

Brassica SEED COATS—*Concluded*

	Surface view	Subepidermal layer (radial)	Palisade cells (radial)
B. nigra (L.) Koch. Fig. 17		Outer row celled	Shallow tumbler cells of even height. Net is caused by the upper limiting palisade walls
B. arvensis (L.) Rabenh. Wild mustard		Outer row celled	Tumbler cells, tall, narrow, of uniform height and crowded closely together
B. alba (L.) Rabenh.	Walls of epidermal layer are collenchymatously thickened	Mucilage appears as in concentric rings around a circular center	

III. Seed and Seedling Pigments*

(Their Importance in Seed Identification)

The four pigment groups that may be utilized in identification of seeds of the genus *Brassica* are the chlorophylls, the anthocyanins, and the carotinoids in the seedlings and the pigments found in the pigment layer or impregnated in the lignified portion of the palisade cells of the seed coat.

The leaf greens of the *chlorophyll* group in both the cotyledons and first seedling leaves are characteristic. The cotyledons of *B. balearica* Pers. and closely related species are very dark blue-green while those of the kales are of characteristic colors. The cotyledons and first seedling leaves of *B. pekinensis* Rupr. are lettuce green, those of colza rapes are bright Rinneman's green, while in *B. nigra* (L.) Koch a typical tint of American green can readily be detected. The practical use of this coloring in identifying seeds may be demonstrated by seedlings grown in the Copenhagen germinator, which, if subject to continuous illumination, will give indication of their inherent cotyledon leaf color within two weeks. The color of the primary leaves is also important in identification of seedlings and is included in the seedling descriptions of Section IV of this paper.

Experimenting with *anthocyanins* has proved that while they have a certain significance, they are not true identification factors. In the first place very few *Brassica* varieties are without white strains. This may be the result of lack of a pigment-forming substance, or the inhibition of pigment formation in the plant. Secondly the amount as well as the intensity of color of the anthocyanins formed in the plant in which they occur is directly dependent on the sugar content of the cell along with the temperature, the moisture supply, and the sunlight to which the plant is subjected. Perhaps low temperature, plenty of direct sunlight, a low moisture supply, and a high sugar

* *The color standard used throughout this work was: Color standards and color nomenclature by R. Ridgway. Washington, D.C. 1912.*

content in the cell were responsible for the very high color attributed to certain *Brassica* seedlings by Muravieva (1928 (41)) and by Orechova and Lapinskaja (1936 (44)). Dorph-Petersen (1927 (11)) finds color in the Swede turnip hypocotyl. Korpinnen (1933 (32)) was unable to detect this color but stated that her seeds were grown inside in poor light. In my experiments it was not noted even when the seedlings grew in sunlight out-of-doors in the autumn, under which conditions some of the kales colored strongly.

The presence of anthocyanins in plants is according to Kajanus (1913 (25)) controlled by simple factors. He states that in the skin of the purple top Swede turnip bulb there is a factor for light purple in the under part of the bulb and one for dark purple in the skin of the upper storage portion. When both are absent the skin is green. In the turnip bulb there are two independent characters determining skin color, *F*, a factor for green skin, and *P*, a factor for red skin. When *F* and *P* are absent the skin is cream yellow and both are independent of each other.

Brassica seeds were germinated between blotters in a dark germinator held at 18° to 20° C. At the end of seven days, as a rule, certain color differences were manifest. These color differences were found to be accentuated by immersing the cotyledons in a 65% solution of lactic acid and leaving them for a time at room temperature. A certain proportion of the anthocyanin pigment is soluble in the cold weak acid and within broad limits will place the plant. Thus considering the *Brassica* species in the order as listed in Section IV of this paper we find purple anthocyanins prevalent in *B. oleracea* L. but diminishing in quantity until in some varieties of *B. Napus* L. no pigments are present. In *B. napobrassica* Mill., which perhaps should precede *B. Napus* L., the color of the acid solution will indicate whether the rutabaga is a purple top, a bronze top, or a green top strain. In *B. Rapa* L. red anthocyanins appear in small quantity, and that generally on injury, but the red pigments become more prevalent in *B. campestris* L. and are decidedly marked in *B. juncea* (L.) Coss. and *B. alba* (L.) Rabenh.

This general knowledge is useful and *could certainly be used by an analyst or grower intimately acquainted with the strains involved*, but definite identification of *Brassica* seeds generally by the use of anthocyanins is dangerous.

Table I gives an indication of the pigments that appear most commonly in the very young *Brassica* seedlings grown in the dark.

The third group of color pigments, the *carotinoids*, is of special importance in *Brassica* distinction. In the cotyledonary leaf of the yellow varieties of *B. napobrassica* Mill., the rutabaga, and *B. Rapa* L., the turnip, are carotinoids not found in any other variety of *Brassica*. They are present in granular form in the plastids in the mesophyll tissue, some portion becoming dissolved in the small oil drops within the cell. Their appearance or absence in the turnip or Swede cotyledonary leaf is governed by the same factors that control flower color and bulb flesh color in these varieties. This will be of great interest to the seed analyst or to anyone such as the geneticist or plant breeder, who appreciates speed in distinguishing very young seedlings.

TABLE I

SEEDLING PIGMENTS

	Anthocyanins				Carotinoids
	Edge of cotyledon	Hypocotyl	On injury	Lactic acid	Cotyledon
Thousand-headed kale	Possible purple tinge	Possible purple or mauve	Purple	Deeply colored mauve or purple	Lemon yellow, some deeper
Green kales			Purple	Purplish-gray or yellow	Lemon yellow, fading
Asparagus kale				Purplish-gray	Greenish-yellow, fading
Collards		Possible purple			
Sheep kale				Colorless	Light yellow
Marrow kale	Possible purple	Possible purple		Deep mauve	Lemon yellow, fading
Ragged Jack kale				Almost creamy	Light yellow, fading
Cabbage and savory cabbage		Purple possible but rare		Yellow or purplish-yellow	Very deep yellow to pale yellow
Brussels sprouts	Mauve or purple	Possible mauve or purple		Slight or deep purple	Deep yellow to lemon yellow
Kohlrabi	Mauve	Purplish-mauve or purple		Slight or deep purple	Pale yellow
Cauliflower	Rarely colored red	Mauve or purple possible		Pale mauve, pink, or pale yellow	Deep yellow to pale yellow
Broccoli	Rarely colored	Rarely colored		Purple or clear light yellow	Deep yellow to pale yellow
B. alboglabra	Reddish-purple			Clear to reddish-purple	Medium yellow to yellow
B. pekinensis	No visible anthocyanins			Clear	Pale to deep yellow
B. Napus	No visible anthocyanins			Yellowish-green	Shades of yellow
B. Napobrassica	No visible anthocyanins			Possible purple or amber	Naples or chrome yellow
B. chinensis	No visible anthocyanins			Amber	Pale yellow
B. rapa	Color very rare		Red	Yellow	Mat or buff yellow or pale yellow, fading to chalky white
B. campestris, annual form	Possible red	Possible red	Red	Clear straw	Light or deep yellow
B. campestris, biennial form	Possible red	Possible red		Clear straw or rose amber, trace of amber	Deep yellow

TABLE I—*Concluded*

SEEDLING PIGMENTS—*Concluded*

	Anthocyanins				Carotinoids
	Edge of cotyledon	Hypocotyl	On injury	Lactic acid	Cotyledon
B. juncea					Lemon yellow
B. arvensis	Possible red				
B. alba	Red	Red		Deep red	Pale yellow

As early as 1900 Percival discovered this color difference. Later Hallquist (1919 (20)) and Nilsson (1924 (42)) considered it. They, however, worked in Europe where white-fleshed Swede turnips are common. In Canada where their presence is extremely rare the importance of this pigment as a distinguishing factor is infinitely greater. Out of at least one hundred samples of Swede turnips examined, I have seen only two samples of white-fleshed Swede turnips and they did not represent commercial seed. In the common turnip, white forms are very common but the turnip seed is much more easily identified than that of the Swede turnip wherein the identification of certain strains is so difficult that for reliability with speed, this color test is the only one I know. Without exception it identifies yellow-fleshed Swedes, which in Canada represent the Swede turnip or rutabaga.

Color inheritance in the turnip and rutabaga group has been investigated and complete association has been established between their flower color and their root flesh color (Malinowski, 1929 (39)). Kajanus (1913 (26)) believes that the corresponding characters of turnips and rutabagas are due to identical factors. In the case of *B. Rapa* L. he suggests a factor *M* for white flesh inhibiting yellow. He found that this white flesh is strictly associated with bright yellow color of flower while yellow-fleshed plants have mat orange flower color.

As regards minor differences, Sylven (1927 (60)) describes four tints of yellow flower color in *B. Napus* L. var. *oleifera* DC. that he uses in the F_1 generation as an index in working out problems of inheritance. These served quite well in the F_1 generation but not with F_2 plants.

McMaster Davey (1931 (38)) notes that flesh color in the turnip is governed by simple factors of which that for white color is dominant with probably minor factors governing the intensity of the yellow. In rutabagas he finds two distinct but similar pairs of factors each of which are capable of producing white flesh color even when several factors for yellow are present. He advises the use of flower color in keeping stock true to type as flower color and flesh color are controlled either by the same hereditary factors or by closely connected hereditary causes. In rutabaga, yellow-fleshed plants have Naples yellow flowers; white-fleshed turnips, white-fleshed rutabagas, and rapes have

bright lemon yellow petals; while yellow-fleshed turnips have mat buff yellow petals, which, however, are more chalky in appearance than their counterpart in rutabagas.

Thus it is agreed that the factors that control flower color in the turnip and rutabaga control flesh color in these plants, and my results of experiments carried on previous to 1932 suggest further that these same factors also determine pigment formation in the cotyledonary leaves.

In the present work on pigments, the seeds of different strains of rutabaga, turnip, and other *Brassicas* were germinated as for anthocyanin pigments and the seedlings immersed in 65% lactic acid and allowed to stand. Carotinoid pigments are not soluble in the acid, but the latter acting on the cotyledonary leaf tissue has the effect of making it transparent (Simpson, 1929 (57)), and thus bringing the leaf pigments into prominence. The yellow rutabaga cotyledons changed to a deep cadmium or Naples yellow while the yellow turnip showed a chalky mat orange color. A solution of methyl orange in water of the strength of 3.9 mgm. to a liter gives a fair indication of the rutabaga seedling color. If any doubt exists, seedlings of 8, 9, or 10 days may be used. After 12 hr. the cotyledonary color begins to fade, in some more quickly than in others. For an indication of carotinoid color in the cotyledons of the different varieties of *Brassica* see Tables I and II.

Through the courtesy of the late R. I. Hamilton, of the Central Experimental Farm, Ottawa, I was enabled to compare directly the color of the flesh of 50 strains of turnips and rutabagas with that of the seedling cotyledons grown from the same seed. The cotyledons were immersed in the acid, as were also sections of the bulb flesh. Comparing them it was noted that a distinct division occurred between the white and yellow turnips. It was also evident that just as the flesh color varies even within types so also does the intensity of the yellow and chrome in the corresponding cotyledons. The roots with intensely colored flesh develop from seedlings with cotyledons colored deep cadmium yellow, those with lighter Naples yellow colored flesh from a paler Naples yellow cotyledon while the white Greystones and Milans have a typical rape yellow cotyledon—the yellow varying in intensity with the depth of (cream) color in the root flesh.

A study of the accompanying Table II may serve to illustrate this.

The actual pigments in the rhizocorm of the rutabaga were investigated by Lubimenko (1914 (37)), who found a small quantity of lycopene as well as an oxidation product of lycopene that he called a lycopinoid. It was more soluble in alcohol and spectroscopically had more intense bands. Tunman and Rosenthaler (1931 (62)) retained the name lycopinoid but Palmer (1922 (46)) and Kylin (1927 (35)) classified under carotinoids both carotene, $C_{40}H_{57}$, and its isomer lycopene, $C_{40}H_{57}$, with the oxidation products of each. In the flesh of the rutabaga Kylin found carotene and its oxidation product xanthophyll and also lycopene and a red carotinoid that is an oxidation product

TABLE II

PIGMENT PECULIARITIES IN SWEDE AND TURNIP VARIETIES

Variety	Number	Shape	Bulb pigments			Seedling pigments	
			Skin		Color of flesh	Flesh immersed in lactic acid	Cotyledons immersed in lactic acid
			Top	Bottom			
Milan Earliest Purple Top	3	Flat, round	Purple	White	Greenish white	White	Palest yellow
Milan Early Purple Top	4	Flat	Purple	White	White		Palest yellow
Purple Top Mammoth Improved	673	Round	Purple	Mauve and white	Pure white	White	Pale yellow
Greystone	711	Flat, round	Purple	White	Pure white	White	Pale yellow
Greystone	12	Flat, round	Purple	Creamy white	Pure white	White	Pale yellow
Red Paragon	682	Round	Purple	White	Pure white	(Darkened)	Pale yellow
Pomeranian White Globe	684	Round	White		White	White	Pale yellow
Southern Prize Turnip	15	Round	Mauve	White	White		Pale yellow
Greystone	10	Oval	Purple	White	White	White	Pale yellow
Greystone	674	Round	Bronze	White	White, cream line just inside skin	White	Pale yellow
Cowhorn	14	Long	Bronze	White	White	Creamy white	Yellow
Improved Greystone	678	Oval	Purple		White	Creamy white	Yellow
Purple Top Milan	2	Flat	Purple	White	White	Creamy white	Yellow
Large White Norfolk	706	Flat	Bronze	White	Slightly creamy white	(Darkened)	Yellow
Imperial Green Globe	692	Round	Green		Slightly creamy white	Creamy white	Yellow
Green Globe	694	Round	Green		Slightly creamy white	White	Yellow
White Globe	705	Round	White		Creamy white	Creamy white	Yellow
Centenary Turnip	688	Oval	Cream yellow		Creamy chrome	Creamy chrome	Chrome
Aberdeen Purple Top	671	Round	Purple	Cream	Creamy chrome streaked with white	Clear chrome yellow	Clear chrome

TABLE II—*Concluded*

PIGMENT PECULIARITIES IN SWEDE AND TURNIP VARIETIES—*Concluded*

Variety	Number	Shape	Bulb pigments			Seedling pigments	
			Skin		Color of flesh	Flesh immersed in lactic acid	Cotyledons immersed in lactic acid
			Top	Bottom			
Aberdeen P.T. Yellow	8		Purple		Creamy chrome streaked with white	Light chrome	Chrome
Early Sheepfold	686	Oval	Green	Yellow	Pure creamy chrome	Chrome	Chrome
Aberdeen Perfection Green Top	695	Round	Green	Chrome-yellow	Spotted chrome and white	Chrome	Chrome
Yellow Tankard	687	Long, oval	Yellow		Chrome	Chrome	Chrome
Yellow Tankard	715	Round	Purple	Yellow	Chrome streaked with white	Deep chrome	Chrome
Golden Vail	716	Round	Green	Yellow	Deep chrome	Chrome	Deep chrome
Aberdeen Green Top	708	Round	Bronze	Yellow	Chrome	Chrome	Deep chrome
Aberdeen Purple Top	710	Round	Bronze	Yellow	Deep chrome streaked with white	Deep chrome	Deep chrome

of lycopene and which he called arumin. To discover whether these same pigments are present in the yellow strains of rutabaga and turnip, the following investigations were made.

The cotyledonary leaves were nipped off and dried in a low temperature oven at 40° C. To 0.8 gm. of this material freshly ground, 5 cc. of a mixture of 90% petroleum ether and 10% ethyl alcohol was added and the pigments were extracted by boiling. The petroleum ether solution was repeatedly fractionated with 98% aqueous methyl alcohol, and then evaporated. A portion tested with chloroform and antimony trichloride gave the blue color indicative of carotene. The Tswett adsorption method as outlined by Schertz (1929 (54)) was tried on the remaining portion but the carotene solution passed through the calcium carbonate column without being adsorbed.

A comparative test of the pigment from the cotyledons of Orange Jelly turnip, Canadian Gem rutabaga, and Dwarf Essex rape with that of the pulp of the Canadian Gem rutabaga rhizocorm was made. To 0.8 gm. of the dried cotyledons, freshly ground, 5 cc. of 95% alcohol was added and allowed to stand at room temperature over night. The fruit of the tomato

known to be rich in lycopene and the root of the carrot, which contains considerable carotene, were used as check tests. These, as well as the turnip root, were cooked in water until soft. The pulp was then filtered, pressed, quickly washed with 95% alcohol, filtered, and again pressed between filter papers. To 0.8 gm. of this pulped material 5 cc. of 98% alcohol was added as in the case of the seedlings. On the second day the alcoholic solutions were filtered off and 5 cc. of fresh alcohol added. This process was repeated on each of four consecutive days. On the fifth day 5 cc. of ethyl ether was substituted for the alcohol. On the sixth day more ethyl ether was added while on the seventh day any pigment remaining was removed by boiling for 10 min. in absolute alcohol plus a small amount of ether, then adding more ether, and allowing to stand over night. Each day the different solutions were standardized by comparison with lead chromate solutions of different intensities. The results are graphically pictured in Fig. 1. As lead chromate

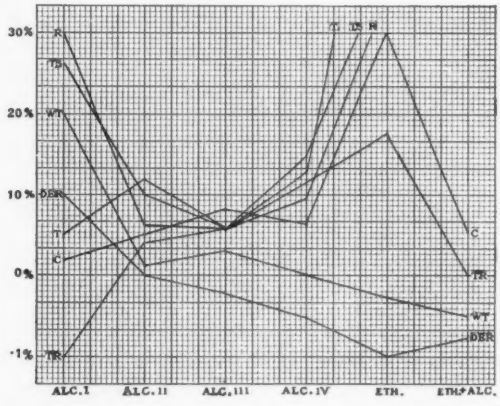

FIG. 1. *Abscissa—solvents used; ordinate—percentage by weight of lead chromate solution used in comparison; R.—Canadian Gem rutabaga seedlings; T.S.—Orange Jelly turnip seedlings; D.E.R.—Dwarf Essex rape seedlings; W.T.—White Milan turnip seedlings; T.R.—turnip root pulp; T.—tomato pulp; C.—carrot pulp.*

only demonstrates yellow grading into chrome, it is quite inadequate where red colorations appear and thus the ether and alcohol solutions of the tomato and the seedlings of the turnip and rutabaga do not appear on the graph. In the first three alcoholic solutions of all cotyledons, xanthophyll appears. There is also present in the first alcoholic solutions of the cotyledons of yellow turnip and rutabaga seedlings some pigment the color of Naples yellow. This is not present in the white turnip or the rape cotyledons. It is readily soluble in carbon disulphide, appreciably soluble in low boiling petrol ether, and in acetone. These properties along with the color would indicate a lycopene-related pigment.

If the ether solutions from the cotyledons of the yellow turnip and rutabaga as well as those from the tomato pulp are concentrated and evaporated on a microscopic slide, crystals are formed. These appear as fine needle-shaped crystals that are found singly or in sheaths. They are pictured in Fig. 2, and have the typical form of lycopene crystals. Crystals may also be procured by extracting the color from the freshly ground cotyledons with carbon disulphide and shaking with petroleum ether. Redissolve the crystals that form in carbon disulphide and again wash with petroleum ether. When the solution is sufficiently concentrated, there appears, on evaporation, an irregular brownish-red spherical crystal mass from which finer crystals radiate as from a central core. These radiating crystals appear needlelike and are also typical lycopene crystals. On the slide from the ether solution besides the lycopene crystals there is another pigment, amber or reddish-brown in color that does not crystallize out of solution. It is dissolved and tightly held in the small fat globules dissolved from the cotyledon and very closely resembles the arumin found in the rutabaga bulb flesh by Kylin (1927 (35)). In an effort to dissociate it the fat from a carbon disulphide solution of the dried cotyledons was saponified with a 20% alcoholic solution of sodium hydroxide and the pigments separated from the soda solution by adding water and ether in a separatory funnel. The other solution was then washed with twice its volume of distilled water and allowed to evaporate on a micro slide. The pigment was evident but crystals were not formed.

Thus we find in the cotyledons of the yellow turnip and of rutabaga seedlings the same pigments that were present in the adult turnip and rutabaga flesh: carotene, xanthophyll, lycopene, and arumin with more of arumin than lycopene.

Perhaps these pigments are controlled by multiple factors and the varying amounts produce the color differences in the cotyledon, in the flower, and in the root bulb flesh but in all probability they are superimposed upon the ground chlorophyll pigments with cumulative effect and are each controlled by individual simple factors.

In any case the cotyledons contain the same pigment as the bulb flesh and are an index of the color of the flower and of the bulb flesh. In consequence they may be used to practical advantage in the distinction of the seeds of yellow-fleshed rutabagas, which in Canada represent the rutabaga, and of yellow-fleshed turnips, by means of the very young seedling.

In Table I, the colors produced by these substances in young cotyledons are listed beside those of the anthocyanin pigments.

The fourth group of pigments are those to be found in the pigment layer of the seed coat or impregnated in the lignified portion of the palisade cells. More general use has been made of this group. For example, if seed coats of *Brassica arvensis* are boiled with chloral hydrate solution and examined under the microscope a red pigment bleeds from the palisade cells.

Brassica seeds change color on ripening and sometimes it is difficult to distinguish mature seeds. Sirks (59) proved that the red seeds commonly found mixed with the dark in certain varieties of rape were not immature seeds but typified a definite strain. Perhaps several *Brassica* seed varieties are mixtures of strains as their color would indicate because they seem to remain true to type, and seed coat color has been found to be one of the most generally reliable of the macroscopic characters of the seed for use in seed distinction.

The presence of carotinoid pigments in the cotyledon forms the basis of the following practical laboratory test.

METHOD for the Separation of Swede Turnip and Dwarf Essex Rape Seeds by Means of the Carotinoid Pigments in the Cotyledon

Germinate the seeds between blotters *in the dark*. It is much easier if they are spaced carefully on the blotters. Allow for seven days' growth *in the dark*, or if the cotyledons have not opened in that time, longer. Fit filter paper in a covered Petri dish and saturate it with a 75% solution of lactic acid, using 15 cc. of solution for a Petri culture dish of 100 mm. diameter. Work quickly. Clip off the cotyledons of the seven-day-old seedlings and transfer them to the soaked filter paper, carefully arranging them in rows. Let stand *in the dark* for 10 or 12 hr. It is convenient to do this work in the afternoon and study the cotyledons the next morning, checking a couple of times through the day. After 24 hr. the cotyledons begin to fade. After the cotyledons have been 10 hr. in the lactic acid, remove the chrome yellow colored cotyledons or those of the Swede turnip to one side of the Petri dish and the lemon yellow or the Dwarf Essex rape cotyledons to the other side and count to find the percentage of each in the mixture.

IV. Descriptions and Keys for the Identification of *Brassica* Seeds and Seedlings

The shape of the seeds of a variety is not always uniform, but on the other hand if the seed is placed in such a way that the raphe is higher than any other portion of the seed and the radicle faces to the left, the characteristic shape or shapes of the species may be noted and in good seed specimens these shapes make up the bulk. Thus *B. oleracea* L. has four typical seed shapes, Figs. 18, 19, and each of its varieties by contributing its own individuality to each of these four shapes gives a clue to its identification. In samples of poor quality, of course, misshapen and diseased seeds require more specific methods. It is also apparent that there is a small percentage of seeds, more especially in strains of cabbage, which do not conform, and as a consequence shape cannot be depended upon as a definite purity test for all seeds.

The size of the seeds is given in three dimensions, the measurements being made with calipers graduated to the sixty-fourth of an inch. The use of this small measurement serves to emphasize minute differences in the comparative size of the different seeds as well as in the different dimensions of the one seed.

PLATE I

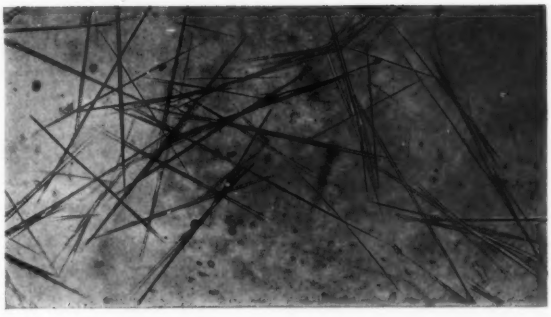

FIG. 2. *Crystals from alcoholic ether solution.*

This measurement was multiplied by 0.4 to obtain the approximate millimeter reading. For the purposes of this record the average of 100 seeds was taken. In a sample of seed the size necessarily varies according to the conditions of growth and the screen over which the sample was cleaned. As the cleaning screen used for a variety of commercial seed is more or less constant, the average measurement of 100 seeds in different commercial samples of a variety varies surprisingly little. It must never be forgotten, however, that seeds of improved strains of a variety are sometimes exceptionally large and also that in any mixture such measurements are useless.

The surface appearance of the seed coat must also be taken into account varying as it does from finely granular to deeply pitted. Even within the species this difference is sufficient to separate two varieties far removed from each other in the series, e.g., cauliflower and marrow kale, but it is disappointing as a means of distinguishing two varieties closely related. Besides, variation within the variety is often too great.

The hilum and the raphe of the seed may often be used to distinguish species and sometimes varieties. In *B. oleracea* L. and *B. Napus* L. the raphe, which appears to be composed chiefly of a bundle of vascular elements, is definitely outside of the hilum, but in *B. Rapa* L. and *B. campestris* L. it overlaps. The hilum in *B. oleracea* L. is large, oval, and rough; in *B. Napus* L. it is large, oval, and flat; in *B. campestris* L. it is distinctly round while in *B. Rapa* L. it is very small.

The cotyledons and radicle of the seed studied when the seed coat is removed are, according to Orechova and Lapinskaja (1936 (44)), useful in identification. There is no doubt that minor differences occur and are evident but after making a determined effort to use this method for identification it was found that these differences were too slight and irregularities in the numerous strains too frequent to make it practical. Cauliflower seeds can be identified in this way although some forms of broccoli are very similar. Brussels sprouts seeds are also typical but there are many irregularities in the cabbage seed. However, as in the case of anthocyanins, any positive characters along this line should be noted for evaluation before proceeding to another method of identification.

When all else fails the seedling plant offers a solution in the shape of the cotyledon and the first seedling leaves, in the presence or absence of an internode, and in the general appearance of the plant. Small pasteboard flats in which the seeds are planted one-half inch apart in a sterilized mixture of two parts loam and one part sand are easily handled and fit neatly into large wooden flats. Special care must be taken in the watering, and sufficient light is necessary. Check tests of closely related seeds are essential.

In the study of seedlings as means of identification of seed, much stress has been laid on the hairs of the leaves, their position, and shape. I have found these very confusing. Hairs on seedling leaves would seem to be affected by the climate, their development being responsive to stimulus perhaps of light, temperature, moisture supply, or other cause. I have found in growing seedlings both under glass and outside that under the conditions

of my experiments the presence of hairs rather than their absence, coupled with inconsistencies in their position and extent, is to be expected. Hellbo (1933 (22)) claims that some hairs on the seedling leaf of turnip are bent at a right angle and may be used as a distinguishing feature. This is quite true of some but not of all strains and doubt arises as to whether in some cases hairs accidentally bent may not be confused with those that inherit this form.

It is also claimed that the rutabaga might be identified by the presence of hairs on the front of the petiole but here again they are not present in all seedlings and they are found on some rape seedling petioles. A certain amount of caution should be used and in any case I do not believe the presence or absence of hairs sufficient to be used alone as a distinguishing feature.

Lavrova (1932 (36)) pictures very young seedlings before the seedling leaves have developed, utilizing cotyledonary shape and inherent seedling behavior of growth. As a matter of fact these two points are extremely important but in such very young seedlings the differences are so slight they are liable to be affected by physical agencies during germination.

The following descriptions apply to seedlings that have formed one or two seedling leaves. They are based on the most general features noted in at least 100 seedlings of every variety available, using the seedlings from seed tested by growing them to mature plants in the greenhouse or garden as the basis of description. Thus the descriptions are composite taken from several hundred plants whereas the illustrations are drawings from photographs of an individual plant chosen because it illustrated, perhaps emphasized, one or more representative points considered important.

For convenience of presentation the arrangement of the groups, in so far as seed character associations have allowed, is that of O. E. Schulz (1919).

The size of cotyledon, the width by the depth, measured in inches (1 in. = 25.4 mm. approx.) is the largest attained by them in the aforementioned growing tests. These were carried on without the use of artificial stimulant.

The size of the seeds was also measured in inches, the sixty-fourth of an inch being the unit used. This may be multiplied by 0.4 to give the approximate millimeter reading. Thus a seed measuring $\frac{3.75}{64} \times \frac{3.5}{64} \times \frac{3.25}{64}$ in. is given as $1.5 \times 1.4 \times 1.3$ mm. The measurements given, taken in three dimensions, are the average of those of 100 seeds of as many varieties as were available.

The colors mentioned refer to Ridgway's color standards (51).

1. **Brassica balearica** Persoon, C. H. Syn. pl. II. 206 (1807); Schulz, O.E., Engler's Pfl. H. 70, IV, 105. 25 (1919)

   Fig. 20. Uncommon in Canada.

   Seed—$2.04 \times 1.84 \times 1.64$ mm.; Natal and bone brown, hazel and chestnut brown, drab and hair brown, and wood brown. The seed is surprisingly similar to that of the common broccoli, in color except for the

absence of the deep olive gray seeds and less of the chestnut brown, in shape except for the absence of the heart-shaped seeds and the presence of a larger number of the broad shallow seeds of the kales, and in seed coat except that the regular shallow oval cavities are just a little coarser. It is generally slightly larger in size.

Cotyledon—1.8 × 1.3 cm.; almost *slate gray* at opening, later becoming *glaucous blue* with reddish edges and veins; thick; translucent; one slightly larger with longer petiole; indentation wide, shallow, and angled at edges.

Seedling—sturdy; stems reddish.

Seedling leaves—dark blue green; shiny; thick; glabrous; veins tinted red at base; ovate, acuminate at base with or without particles down stem; margin irregularly incised; surface undulate.

2. **Brassica macrocarpa** Gussoni, G., Ind. Sem. Hort. Reg. Boccadifalc 3 (1824–5); Schulz, l.c. 26

Fig. 21. Uncommon in Canada.

Seed—2.0 × 1.8 × 1.5 mm.; hair brown, Natal and bone brown; resembling thousand-headed kale in shape but lacking the broad shallow seed of the kales; the seed coat has a regular, fine, medium deep stippling.

Cotyledon—0.79 × 0.95 cm.; *dark green* with lighter colored veins tinted purplish-red at base; shiny; thick; heart-shaped and continuous with the petiole in a graceful sweeping curve; *indentation* narrow, deep, and arcuate; distinguished from *B. oleracea* L. by its color.

Seedling—fine; petiole green, tinted purplish-red.

Seedling leaves—green covered with a white glaucous coat; thin; glabrous; oblong to ovate, acuminate at base; margin irregularly but somewhat biserrately incised; surface undulate.

3. **Brassica oleracea** L. Spec. Pl. Ed. i.II.667 (1753)

Distinctive features of the species:

(i) Seed color gray and brown predominating (see seed key); seed size varying (see text), and seed shape typical (see Figs. 18 and 19). The raphe is always outside of the hilum, which is large, oval, and rough. The micropyle is above the usually raised tip of the radicle ridge.

(ii) Radial section of the seed coat (Fig. 4).

(iii) Purple anthocyanins are common to the colored strains of the species but are lacking in the white strains. Table I.

(iv) Seedlings have a glaucous coat, varying slightly in color and intensity according to variety but typical and easily recognized.

A. *Brassica oleracea* L. var. *ramosa* (DC). Alefeld., Landw. Fl. 234 (1866); Schulz, l.c. 30

B. arborea Steudel, E.G., Nom. Bot. ed. i. 116 (1821)

B. oleracea L. *B. acephala* DC. subvar. *ramosa* DeCandolle, O.P., Syst. Nat. II. 583 (1821) et. Prodr. 1. 243 (1824)

Bush kales, branching kales.

I. Flanders kale. Fig. 22. Not common in Canada.

Seed—1.8 × 1.76 × 1.6 mm.; olive gray, olive and clove brown, fuscous; majority of seeds are quite as wide as high but a few of the tall type of seed as found in thousand-headed kale are present, seeds often come to rest with the hilum and the micropyle visible; seed coat has an extremely shallow primary net and a conspicuous secondary net, and is generally fine with buff-colored wax in patches on the surface.

Cotyledon—1.1 × 0.79 cm.; often three cotyledons of deep dull yellow-green; shiny; thick; triangular; *indentation* very broad, shallow, and rounded.

Seedling—strong; slow growing; short; stem green; hypocotyl tinged with red.

Seedling leaves—slightly glaucous; oval, truncate tip and base; coarsely, irregularly, and deeply serrate with four serrations on each side.

II. Thousand-headed kale. Fig. 23. Extensively grown in Canada.

Seed—Fig. 18; 2.04 × 1.8 × 1.64 mm.; olive gray, olive and clove brown; rough, viewed from above elongated and slightly *wedge-shaped*; very heavily coated with *buff-colored wax* some of which chips off; primary net in the seed coat, angled, its walls shallow, narrow, and perpendicular; secondary net. appears in horizontal rows across seed; seed coat very thick.

Cotyledon—very small in some strains but may reach 1.27 × 0.95 cm.; chromium green; thick but thinner than broccoli; one larger and kidney-shaped, other inclined to be heart-shaped but both have flared bases; closely resembles sprouting broccoli in shape but more like cabbage in texture and in not bending at right angles to the petiole; indentation medium broad, deep, and rounded.

Seedling—irregular; sturdy; tall; stem thick sometimes purplish at base; slower growing than broccoli. Table I.

Seedling leaves—*glaucous gray above and below* with trace of purple at base; thick with veins prominent although not so deeply sunken as in marrow kale; glabrous; obovate to cuneate; shallowly sometimes sharply serrate and biserrate.

B. *Brassica oleracea* L. var. *acephala* DC. l.c. 583; Schulz l.c. 30

Leaf kales—grown only to a limited extent in Canada.

I. Kitchen kales. They are variously classified according to color, habit of growth, and the degree of laciniation of the leaf.

a. Tall green curled, or Scotch kale; intermediate moss curled kale; dwarf green curled or Labrador kale. Fig. 24.

Seed—1.9 × 1.72 × 1.64 mm.; *clay color* with seeds of *tawny olive* conspicuously present; primary net extremely shallow *on* the seed coat.

Cotyledon—1.43 × 1.1 cm.; light bice green later becoming darker; dull; deep central vein; indentation circular and deep, broader in one than the other.

Seedling—fine; slender; tall green kale is a taller seedling and has a narrower indentation in the cotyledon; *stem petiole* and *median vein* a characteristic *apple green.* Table I.

Seedling leaves—bice green slightly blue glaucous above and slightly gray glaucous beneath; thin; dull; upright; usually glabrous but may have setose hairs on the edges; appearing early and early showing the undulate pinnasecti character of the leaf.

b. Asparagus kale or Jerusalem green curled kale. Fig. 25 *b.* Grown in Canada but not common. Asparagus and ragged Jack kales have been placed by many writers along with the winter and oil rapes, *B. Napus* L. While these seeds resemble *B. Napus* L., Fig. 6, in not having a celled outer epidermal row in radial section of the seed coat, they have the tumbler cells with narrow straight-sided walls of uniform height, flattened above and below, so typical of *B. oleracea* L. varieties, Fig. 4. They are without doubt intermediate forms.

Seeds—1.76 × 1.7 × 1.6 mm.; shaped like the rape they normally rest with the hilum above; the hilum, raphe, and micropyle are also similar to those of the rape in size, shape, and placement. However, the mucilage that often covers the hilum and that turns brown on old seeds, the blue gray color of the seed, and the large rectangular meshed waxed net in the seed coat are oleraceous characters.

Cotyledons—9.5 × 6.3 mm.; green; not glaucous; thick; indentation small, rectangular.

Seedling—purple hypocotyl, Table I, slow growing; slender; tall; medium green; stem dark brownish-green.

Seedling leaves—Rinneman's green; not glaucous; thin; may have setose hairs on under veins, edges, and petioles; broadly ovate with truncate tip so that it is quite as broad near to the tip as at the center, but as the leaf gets older it becomes almost round; unevenly dentately serrate.

c. Variegated kale. Fig. 28 *c.* A garnishing kale, grown more extensively in Canada than some other strains. Sometimes mixed with ribbon kale.

Seed—1.86 × 1.67 × 1.59 mm.; shapes more nearly resemble those of collards but seeds are more angular and have no raised raphe; shallow forms predominate; net with fine, shallow, squared mesh, of uniform size, and without pattern in the seed coat.

Cotyledons—similar to those of the green kales.

Seedling—opens like that of marrow kale.

Seedling leaves—similar to those of the green kales except for the deep red and blue *anthocyanin* coloring in the lamina. Also they are more ovate and not so deeply incised.

d. Blue curled Siberian kale. A dwarf kale; quite commonly grown.

Seed—1.76 × 1.68 × 1.6 mm.; deep dark mouse gray; somewhat spherical; primary net is distinct but so shallow as to approach in appearance that of *B. Napus* L.

Cotyledons—small; glaucous; asphodel green; dull; thick; heavy; dark purplish-red median vein; indentation broadly triangular.

Seedling—slender; stem and petiole purplish-red.

Seedling leaves—blue glaucous; bristly setose on upper lamina, edges, and petioles; edges deeply cut.

e. Collards. Fig. 26. Plant cut for food in the rosette stage but if left will form a head. Not common in Canada.

Seed—1.84 × 1.72 × 1.2 mm.; Fig. 18; viewed from above a rounded oval; primary net shallow; small; fairly regular; rectangular.

Cotyledon—9.5 × 4.76 mm.; lamina ribbon down stem; deep dull yellow-green; thick; when young sharply angled with the petiole but later straightening; indentation arcuate.

Seedling—gracefully fine; stem greenish-white or purplish-red.

Seedling leaves—shiny; thick; glabrous; veins broad and white; oval or broadly elliptical with obtuse tip and acuminate base; sharply and shallowly serrate.

f. White ribbon kale. Fig. 27 *a.* Mature leaves cut in ribbons but plant forms a head in the fall. Often grown for ornamental purposes.

Seed—1.96 × 1.88 × 1.64 mm.; *mouse gray*; heavily coated with light brown mucilage; primary net very shallow in vertical rows down the rectangular side of the seed; viewed from above ovate.

Cotyledon—9.5 × 7.9 mm.; *white green*; thin; dull; one erect the other forming an angle with the petiole; lamina ribbon for short distance down stem; indentation narrow and deep.

Seedling—small; opening like marrow kale; petiole with red tinge.

Seedling leaves—*white green*; glabrous; veins deep, sometimes purplish; oval with tip and base slightly acuminate but so deeply cut as to give the impression of lobing.

II. Borecoles and kales. Used for the most part as stock food.

a. Sheep kale, kale rape. Grown in Canada in limited quantity.

Seed—1.48 × 1.44 × 1.44 mm.; regularly *spherical*; uniformly deep mouse gray.

Cotyledon—1.27 × 1.59 mm.; dark green; purplish petiole; shallow, broad, rounded indentation.

Seedling—very small; oleraceous. Table I.

Seedling leaves—deep set veins in a smooth surface; hirsute on edges and on under lamina; purplish tint in veins, stem, pedicel, and leaf edge.

b. Cottagers kale. Fig. 27*b*. Uncommon in Canada.

Seed—1.96 × 1.76 × 1.6 mm.; all shades of drab and cinnamon drab; coated with bronze mucilage; viewed from above, rectangular and from the side, the lower left corner is cut away; net small-meshed and shallow, *juncea*-like.

Cotyledons—1.1 × 0.8 × 1.1 cm.; one being narrower than the other; broadly heart-shaped; light green; thick median vein; indentation narrow and triangular.

Seedling—tall and slender; purplish stem.

Seedling leaves—slightly glaucous; shiny; glabrous, orbicular to peltate; more or less dentately serrate or biserrate.

c. Milan kale. Fig. 28 *b*. Uncommon. Fleshy shoots are cut in the spring and later a small head forms at the top of the plant.

Seed—1.8 × 1.8 × 1.8 mm.; tending to *cubical*; shades of drab and olive brown; a net minutely shallow, of small rounded mesh.

Cotyledons—7.9 × 9.5 mm.; broadly heart-shaped, attenuate at base and continuing down stem; indentation deep, angular.

Seedling—tall; slender; graceful.

Seedling leaves—glabrous not glaucous; unsymmetrical; oval; acute tip and cuneate base; secondary leaf with a more truncate tip and more biserrately cut.

d. Marrow kale. Fig. 29. A thick stemmed variety extensively grown.

Seed—2.7 × 1.96 × 2.2 mm.; Fig. 18; dark olive buff, mouse and olive gray, olive and clove brown; dotted with white mucilage; viewed from above wedge-shaped to oval and round; seed coat very *thick*; primary net deep; mesh small with slanting walls.

Cotyledon—1.1 × 1.27 cm.; medium dark green; thin; veins white or purple; *cotyledon completely circular* if the curved irregular indentation and the narrowing of the lamina down the stem are ignored.

Seedling—on opening, the upper larger cotyledon has its edges loosely rolled inward to form a semicircle wholly imbricated within the lower curved smaller one. As the inner elongates the outer remains slightly curved. They then open by separating on one side only; slender; tall. Table I. Stem and under veins white with purple, or green with bronze.

Seedling leaves—light bluish-green; not glaucous, unsymmetrically and deeply serrate or triserrate.

e. Ragged Jack kale*. Fig. 25 *a*. A very common impurity in samples of seed received from southern Europe.

Seed—1.88 × 1.68 × 1.6 mm.; deep mouse gray; almost spherical.

Cotyledons—6.35 × 7.9 mm.; very dark green; the upper when young at right angles to an erect petiole; hypocotyl on germinating curling in an almost complete circle.

Seedling—sturdy. Table I.

Seedling leaves—glabrous; *three- to four-parted with each part* again incised.

C. *Brassica oleracea* L. var. *bullata* DC. subvar. *palmifolia* DC. l.c. 584

Palm tree kale. Fig. 28 *a*. Uncommon, often used for ornamental purposes but one variety in South America is used for food.

Seed—2 × 1.6 × 1.6 mm.; viewed from above round, from the side resembling the rounded and taller forms of savoy cabbage as pictured in Fig. 19.

Cotyledon—9.5 × 7.9 mm.; one erect, heart-shaped, other angled with petiole, much broader; indentation shallow; curved.

Seedling—*bice green stem and petiole* (cf. green kales).

Seedling leaves—somewhat glaucous; shiny; wrinkled; slightly setose on under lamina; obovate sometimes with cuneate base; the primary having a blunt tip; shallowly and crenately dentate.

D. Brassica oleracea L. var. *sabauda* (L.) Martens in Mart. et Kemmler, Fl. Wurttemb. ed. 2. 35 (1865); Hegi, R. Illustr. Fl. Mitt. Eur. IV. i. 246 (1917)

B. oleracea L. var. *bullata* DC. subvar. *sabauda* (L.) DC.; Schulz l.c. 31

B. sabauda Dod. Pempt. 624 (1583) emended (1616); Schulz l.c. 31

Savoy cabbage, Fig. 30. Common vegetable.

* *See asparagus kale.*

Seed—1.96 × 1.8 × 1.64 mm.; Fig. 19; vinaceous drab, pecan brown, Rood's brown, fuscous and fuscous black; viewed from above rounded; seed coat thin; smooth; net very shallow.

Cotyledon—9.5 × 5.9 mm.; the large cotyledon turned back at right angles to its tall petiole overlapping to about one-half of the small cotyledon turned back at right angles to its short petiole; puffed in center as a *balloon top*, Fig. 30; chromium green; shiny; thin; sides rounded; lamina continuing down stem; indentation irregular.

Seedling—tall; slender; graceful; lacking the stiffness of the ordinary cabbage seedling; stem light bice green.

Seedling leaves—dark greenish glaucous; smooth; dull; glabrous; veins sunken; oblanceolate to oval; edges of lamina at center of base tending to curve inwards and form a funnel.

E. *Brassica oleracea* L. var. *gemmifera* (DC.) Zenker. Fl. Theuringen XV. 2 (1824); Bailey l.c. 226

B. oleracea L. var. *bullata* DC. subvar. *gemmifera* (L.) DC.; Schulz 31

B. gemmifera Léveillé Monde. Plant XII. 24 N.V. (1910)

Brussells sprouts. Fig. 31. Common vegetable.

Seed—1.96 × 1.76 × 1.52 mm.; Fig. 18; Saccardo's umber and sepia, russet and hazel; viewed from above rounded; seed coat thin, smooth. Figs. 3, 4.

Cotyledon—1.43 × 1.1 cm.; chromium green becoming lighter; smooth; shiny; on germination the hypocotyl almost perpendicular and in line with the petiole and the two cotyledons also standing *perpendicularly with ventral faces together*; larger cotyledon with broader indentation and more acuminate base.

Seedling—short; stout; hypocotyl, under veins, petiole, and stem green, with bronze or purple, according to variety.

Seedling leaves—Rinneman's green; smooth; glossy; glabrous; primary leaf oval, practically *entire* on a short *erect* petiole. Table I.

F. *Brassica oleracea* L. var. *capitata* L. Spec. Pl. 667, ed. I, II (1753)

Cabbage. Fig. 32. Common vegetable.

Seed—2 × 1.84 × 1.68 mm.; improved varieties much larger; Fig. 19; testaceous, cocoa brown, walnut brown, burnt umber, deep brownish drab, fuscous, and a few deep olive gray; noticeable ridge in upper left side caused by an enlarged radicle tip emerging from between the cotyledons in the seed halfway up the side; irregular, more often wider than deep; primary net, with narrow, straight-sided walls and of large squared mesh, in regular rows.

Cotyledon—**14.2** × 7.9 mm.; in germination the smaller cotyledon opening immediately on being pushed above the soil, the larger one becoming conduplicate, ventral side within, for some time before moving slightly to one side, bending back, and opening away from the other on the opposite side; chromium green; shiny; thin; kidney-shaped; heart-shaped in some strains; puffed at center as in savoy cabbage; indentation arcuate.

Seedling—very sturdy; stem and petiole thick and white with a possible trace of purple. Table I.

Seedling leaves—*funnel-shaped at base*; smooth; dull; pronounced deep greenish glaucous; on sprouting, the primary leaf, conduplicate with ventral side within, is extended at right angles to the stem by the growing petiole, which later straightens to a vertical position and holds the *stiff primary leaf turgidly erect*; broadly oblanceolate to oval with obtuse-angled tip but varying according to strain; shallowly, evenly, and finely biserrate.

G. Brassica oleracea L. var. *botrytis* L.

I. *Brassica oleracea* L. var. *botrytis* L. subvar. *cymosa* Duchesne, Lamarck, Encycl. I. 745 (1783); (*asparagoides* DC. l.c. 586 (1821)

B. oleracea L. var. *italica* Plenck, Plant Medic. VI, 29.t.534 (1794)

B. asparagoides Calwer, Deutochl. Feld. u. Gartengew. 183. t. 28 (1852) Bailey l.c. 227.

Broccoli. Fig. 34. As a vegetable in greatly increasing demand.

1. Types with small heads forming in the axils of the leaves: Italian green sprouting, Calabrese, Propogena.

2. Types resembling cauliflower only maturing later: Walcheren, Early Roscoff.

Seed—1.76 × 1.72 × 1.6 mm.; Fig. 19; hazel and chestnut brown, Natal and bone brown, few deep olive gray and wood brown; viewed from above rectangular and from the side an elongated oval flattened on upper right corner; seed coat resembling cauliflower with the exception of the deeper net and the mesh four times as large.

Cotyledon—deep dull yellow-green; blue glaucous; shiny; thin; although thicker than thousand-headed kale; in Type 2, resembling that of cauliflower, though slightly less angled, but in Type 1, *resembling those of thousand-headed kale*; indentation broad, shallow, and more rounded than that of the kale.

Seedling—tall; slender at first; *slow growing*; stem greenish-white tinged at the base with purple and bronze, Table I.

Seedling leaves—*deep greenish glaucous*; glabrous; smooth; primary leaf, when sprouting, slender, conduplicate, ventral side within, and inverted by a graceful curve in the petiole; ovate with obtuse or acute tip; unsymmetrical; crenately biserrate.

II. *Brassica oleracea* var. *botrytis* L. subvar. *cauliflora* (Gars.) DC. l.c. 586; Schulz. l.c. 33

B. cauliflora Garsault, Explic. Sept. Cents. dix-neuf pl. 123 (1765)

B. botrytis Mill. Gard. Dict. Ed. 8 (1768); Bailey l.c. 229

Cauliflower. Fig. 35.

Seed—2.16 × 1.72 × 1.68 mm.; Fig. 19; drab and hair brown, Natal and bone brown; viewed from above rounded, from the side elongated; coat *fine*; *smooth*; thin; with an exceptionally shallow net, square-meshed.

Cotyledon—*blue glaucous leaf green*; thick; translucent; triangular in shape with apex toward the petiole; about 1.5 mm. from this apex bending at a sharp right angle to the perpendicular stem (Fig. 35 *b*) to form a *flat symmetrical table, rectangular* 2.2 × 1.58 cm., except for the clearly defined angular incisions in each of the four sides (Fig. 35 *c*).

Seedling—sturdy; slow growing; stem, petiole, hypocotyl, and under veins purplish or greenish-white according to variety; petioles elongate greatly. Table I.

Seedling leaves—erect; *deeply greenish glaucous*; dull; glabrous; oblanceolate; finely and very shallowly serrate.

H. *Brassica oleracea* L. var. *gongylodes* L. Spec. Pl. 667 (1753); Kerner, oekon, pfl. IV. 6 t. 344 (1791); Schulz l.c. 32

B. oleracea L. var. *caulorapa* DC. *α communis* DC. l.c. II. 586

B. caulorapa Pasq. Cat. Ort. Bot. Napoli 17 (1867); Bailey l.c. 232

Kohlrabi. Fig. 33. Common vegetable.

Seed—2.2 × 1.92 × 1.8 mm.; Fig. 19; but varying from 1.78 mm. to 2.18; sorghum brown, Hay's brown, and light seal brown, brownish drab, dusky drab, and blackish-brown with an occasional seed of Morocco red and of claret brown; primary net extremely shallow, honeycomb-meshed, with narrow slanting walls often capped with blue-gray mucilage.

Cotyledon—1.58 × 9.5 mm.; chromium green; shiny; glaucous; short lamina ribbon down stem; indentation *broader in one than in other.*

Seedling—*very slender, tall, and graceful*; stem, hypocotyl, and petiole showing traces of vinaceous purple, Table I.

Seedling leaves—dark bluish glaucous; thick, only central vein showing; oblong lanceolate; stands erect on a *tall graceful petiole*; four serrations on one side, three on the other; end lobe elongated.

4. **Brassica alboglabra** Bailey, Gent. Herb. i. 79. Fig. 29 (1922)

Brassica oleracea (L.) Burkill, Gard. Bul. Str. Settlement V. 3–6 (1930)

Chinese kale, guy lon, sawi hijau tuah. Fig. 36. Seed and green plants in season commonly sold by Chinese merchants. The large bundles of stout short stems are conspicuous because of the large white flowers often present. Used for soups, salads, and chop suey.

Seed—2.12 × 1.92 × 1.72 mm.; deep, dark, and blackish mouse gray; viewed from above circular, from the side shield-shaped rounded at the top; net shallow, of medium large square mesh with narrow walls; sparsely spotted with gray mucilage.

Cotyledon—1.43 cm. × 9.5 mm.; turtle green; dull; thick.

Seedling—conspicuously oleraceous; stem tender, white with light purplish tint. Table I.

Seedling leaves—*olivine green*; decidedly glaucous; setose; broadly obovate to round with truncate tip; veins white and large; unsymmetrical; sharply biserrate.

5. **Brassica Robertiana** Gay. Ann. Sc. Nat. i. VII. 416 (1826)

Brassica Montana Pourret, Chlor. Narb. n. 180. m. Mem. Acad. Toul. i. III. 308 (1788); Schulz l.c. 33

Fig. 37. Uncommon.

Seed—1.88 × 1.8 × 1.48 mm.; russet and hazel, Saccardo's umber and sepia; almost spherical somewhat resembling seeds of cauliflower in shape but lacking its graceful spherical curves; seed coat thin; primary net of squared mesh, fine and irregular.

Cotyledon—1.1 × 1.1 cm.; deep, slate green when young, later becoming lighter always blended with bright, purplish red; dull; waxy; heart-shaped with a slight angle at the sides; lamina continuing down stem for a short distance; indentation the shape of the two end lobes of the cotyledon inverted.

Seedling—large; sturdy; stem and veins purplish-red; petioles long and glaucous gray.

Seedling leaves—dark greenish glaucous; glabrous; oval to ovate later elongating slightly; unevenly bidentate; unsymmetrical.

6. **Brassica insularis** Moris, Fl. Sard. I. t. XI. 168 (1837)

Fig. 38 *a*. Not commonly grown.

Seed—1.8 × 2 × 1.36 mm.; Chaetura drab, mars brown, russet brown, heavily spotted with brownish gray mucilage; viewed from above rectangular, from the side triangular with the top and right sides forming a right angle while the third side is arcuate; higher than broad; medium fine net, irregular.

Cotyledons—1.1 cm. × 7.9 mm.; *asphodel green*; medium thin; base red; sides rounded; indentation broad, very shallow and arcuate.

Seedling—large; stems red, petioles elongated.

Seedling leaves—*deep greenish glaucous*; glabrous; large; broadly oval to obovate; undulate, sharply dentate.

7. **Brassica cretica** Lam. Encycl. 747 (1783); Schulz l.c. 36

Fig. 39. Uncommon in Canada.

Seed—light grayish-brown; viewed from above round, from the side oval; primary net fine and indistinct in rows in the seed coat.

Cotyledons—1.4 × 1.1 cm.; deep slate gray; dull; waxy; thick; veins purplish-red; indentation narrow; deep and curved.

Seedling—large; *glaucous green*; stems purplish-red.

Seedling leaves—glabrous; dark green; blue glaucous; dull; veins purplish-red, sunken; broadly oval, more or less truncate at tip and base; quickly enlarging; lamina rough; repand; shallowly and crenately biserrate.

8. **Brassica incana** Tenore, Prodr. Fl. Napol I. i. (1811–15) XXXIX, et IV, Syll.t. 165 (1830); Schulz l.c. 37

Fig. 38 *b*. Uncommon in Canada.

Seeds—large; spherical but slightly elongated; cocoa brown, walnut brown, burnt umber, deep brownish-drab; primary net clear-cut, medium fine and irregular, secondary net fine.

Cotyledon—1.58 × 1.1 cm.; *almost slate gray* remaining dark; dull; thick; translucent; waxy; veins reddish; heart-shaped; indentation shallow and narrow.

Seedling—oleraceous in appearance; *stem hoary gray.*

Seedling leaves—very dark green; glaucous blue above, hoary gray beneath; dull; broadly oval with truncate tip; shallowly and irregularly biserrate.

9. **Brassica pekinensis** Ruprecht, Fl. Ingr. I. 96 in textu (1860); Bailey l.c. 250

B. cernua (Thunb.) Forbes & Hemsley, J. Linn. Soc. XXIII, 47 (1886); Schulz l.c. 59

B. campestris L. sub. sp. *chinensis* L. var. *pekinensis* Burkill.

Pe-tsai, chu toy, celery cabbage. Pe-tsai is a general term but is often commercially associated with the open-headed type of plant. Its use as a vegetable is gradually increasing.

General characters of the species:

1. Seed—1.9 × 1.76 × 1.56 mm. with a wide range of size; vinaceous; viewed from above wedge-shaped, thickest at radicle, from the side circular with an elbow one-third from the top on the right side with the lower right side cut away; net shallow, round-meshed and small; raphe, round and quite the largest of the Brassicas, on the base of the hilum, both enclosed by a second ring.
2. Distinguished from *B. Napus* L. by the brownish-yellow seed coat, sloughed off on germination, that contrasts with the very dark one of the rapes—a color difference not apparent in the dry seed.
3. Seedling stem succulent; white with purple tint; Table I.
4. Shape of seedling leaves is an index to the shape of the mature head.

The following six varieties are grown:

A. Chihili or improved Peking. Fig. 41.

Head large, hard, projectile-shaped.

Seed—1.8 × 1.66 × 1.5 mm.; light and dark vinaceous drab and dark grayish-brown, vinaceous rufus and liver brown, and a few neutral gray; more elongated than other varieties; *oval bodies* in pigment layer of the seed coat; net small and very shallow; secondary net visible.

Cotyledons—1.43 × 1.27 cm.; dull apple green becoming bice green; indentation narrow, deep, and angular.

Seedling—tall; slender; stem purplish.

Seedling leaves—*rapidly elongating*; long hairs on edges, shorter on surface, under veins and petioles; veins prominent, white, broad at base; *oblanceolate* with lamina ribbon down stem; finely and sharply biserrate.

B. Wong bok. Fig. 40.

Head half-long; large; tightly wrapped.

Seeds—1.8 × 1.76 × 1.7 mm.; brownish-drab and dusky drab, hazel and chestnut brown, cinnamon rufus; elbow low and prominent; coarsest primary net of group; secondary net distinct, fine.

Cotyledons—1.44 × 1.1 cm.; lamina ribbon down stem; turtle green; indentation shallow, arcuate.

Seedling—short; stout; stem and petiole thick.

Seedling leaves—decidedly setose; ***round*** when young later broadly obovate, acuminate at base; veins prominent; coarsely and shallowly dentate and bidentate.

C. Chefoo. Fig. 42.

Head—large; round; solid.

Seed—1.9 × 1.79 × 1.55 mm.; dark and light, vinaceous drab, dark grayish-brown, and neutral gray; seed angular; very shallow and flattened primary net.

Cotyledons—1.76 × 1.6 cm.; dull; medium thick; large; veins prominent; indentation broad, circular.

Seedling—large.

Seedling leaves—densely setose; thin; dull; slightly wrinkled; obovate; becoming more acuminate; unsymmetrical, shallowly dentate.

D. Market Pride. Fig. 43.

Head—large, tender, loosely folded like a cabbage.

Seed—1.87 × 1.8 × 1.7 mm.; brownish- and dusky drab, Hay's russet, liver brown and deep mouse gray; prominent elbow; primary net slightly smaller than in wong bok, secondary net distinct.

Cotyledons—1.43 × 1.27 cm.; thin; light bice green; appearing almost rectangular; indentation shallow; angular.

Seedling—*stalky.*

Seedling leaves—sparsely setose; wrinkled; veins broad, white; round becoming oval acuminate at base; almost entire but coarsely and shallowly dentate and bidentate.

E. Shantung.

Head—large; loose and open.

Seed—1.79 × 1.67 × 1.44 mm.; dark and light vinaceous drab; dark grayish-brown, few vinaceous rufus and neutral gray; side view rounded; primary net like chihili but deeper.

Cotyledons—1.27 cm. × 7.9 mm.; veins prominent for short distances; indentation broad and angular.

Seedling—medium coarse; short petioles.

Seedling leaves—purplish glaucous on under side; dull; not wrinkled; veins prominent; setose on edges; spatulate to obovate with acuminate base; almost entire but shallowly serrate.

F. Chokurei.

Head—solid for over three-quarters of its length but from there the leaves flare like lettuce.

Seed—1.8 × 1.8 × 1.56 mm.; resembling wong bok in color but has very fine primary net; conspicuous micropyle and high elbow.

Cotyledons—1.59 × 1.8 cm.; lamina angled at base of side; light bice green; indentation angular.

Seedling—low and medium stout; stem and veins white with mauve tint.

Seedling leaves—shiny; smooth; densely hirsute on edges and under veins but sparsely on lamina; elliptical slightly acuminate at base; long petioles; sharply denticulate.

10. **Brassica Napus** L. Spec. Pl. ed. i. II. 666. n. 3 (1753)

A. *Brassica Napus* L. var. *arvensis* (Lam.) Thellung f. *biennis* (Schubl. et Mart.) Thellung, Hegi. l.c. 254

B. Napus L. var. *biennis* (Schubl. et Mart.) Reichb. Deutschl. Fl. I. 107. t. 93 (1837–38); Schulz l.c. 44

Giant, broad-leaved, or dwarf rapes. Figs. 5, 6. Grown in Canada for pasture, silage, and cover crop. Produces seed the second year but very liable to winter killing.

Dwarf Essex rape. Figs. 45 and 47 *a*. Extensively grown.

Seed—1.84 × 1.76 × 1.76 mm. *with range to 2.12 mm.*; naturally rests with hilum visible; shiny; purplish-gray, dusky and dark neutral gray with some claret brown; viewed from above, smooth, flattened, broadly oval with flattened radicle side; black micropyle; hilum large and broadly oval with cells distinct from seed coat netting; raphe small, outside the clear-cut hilum line but connected with it by two threads of mucilage that circle the central scar in the hilum; from the side, shield-shaped with a wedge-shaped slice cut from the right top; seed coat reticulated; primary net shallow, elongated longitudinally; secondary net broadly rectangular, regular.

Cotyledon—9.5 mm. × 1.43 cm.; deep dull yellow-green; dull; thick; *translucent*; angled with petioles, one of which is longer than the other; indentation broad, but narrowly arcuate. Base also narrowly and shallowly arcuate on each side of petiole; lamina ribbon down petiole.

Seedling—strong; internode; no anthocyanins; Table I.

Seedling leaves—turtle green; blue pruinose; smooth; sparsely hirsute on edges, veins, and lamina; hairs on lamina set in small cushionlike projections; shield-shaped base with end lobe as a flattened dome above the shield; four lobes on each side that may be serrations, bi- or triserrations and basal one often only a fragment down petiole; secondary leaf more rectangular.

B. *Brassica Napus* L. var. *arvensis* (Lam.) Thellung f. *annua* (Schubl. et Mart.) Thellung, Hegi l.c. 254

I. Colza rape, winter rape, oil seed winter common. Figs. 44 and 47 *b*, *c*. In warm countries winter rape is sown in the fall, and plants, after supplying winter pasture, produce seed in early summer.

The term "Colza" at one time was synonymous with *B. campestris* (L.), probably because these seeds were commonly used in oil production. Now, according to Passerina (1935 (47)) the term is attributed to salad oils and the seeds of the *Cruciferae* that produce them whether they be *Brassica*, *Sinapis*, *Eruca*, or *Camelina*.

Seed—1.92 × 1.84 × 1.8 mm.; *dark or blackish mouse gray*; spherical; normally rests with the large flattened oval hilum visible; raphe outside but from it three strands of mucilage fan over the hilum; top of seed not so smoothly flattened as biennial rape, often depressed at micropyle; palisade cells more nearly the same height than in biennial form and primary net is shallow; mesh squared or elongated longitudinally; seed coat netting continuous inside the outer hilum ridge.

Cotyledon—1.59 cm. × 7.9 mm.; deep dull yellow-green; translucent; kidney-shaped; bent at a slight angle to the stiff petiole slightly back from its junction with it; two petioles almost the same length; base broadly arcuate on each side of petiole so that in shape cotyledon appears as two circles touching at midvein; indentation broadly arcuate. This typical cotyledon is not so marked in annual oil seed rape of South American origin grown in Canada.

Seedling—fine despite large cotyledon; stems white; internode.

Seedling leaves—typically *bright* Rinneman's green; blue pruinose; very sparsely setose on edges and veins; veins slightly sunken; either three or four lobes on each side or three on one and four on the other, each serrate or bi- or triserrate; no segments down stem; dome of leaf often elongated.

II. Small-seeded oil rape.

Seed—1.75 × 1.75 × 1.5 mm.; uniform blackish mouse gray; smooth; viewed from above, oval, flattened at radicle; from the side, broad, shield-shaped, very slightly curved on top and of equal width and height; rounded at base and only slightly flattened at upper right corner; primary net fine with very shallow round mesh.

Cotyledons—large, resembling Colza rape.

Seedling leaves—Rinneman's green; thick; large; smooth, very sparsely setose on edges, veins, and under petiole; primary leaf square; secondary square to oblong; large with an extremely shallow broad dome; lamina down stem in shallow wings; unevenly biserrate.

C. *Brassica Napus* L. var. *quadrivalvis* Hook. fils & Thoms. J. Proc. Linn. Soc. Bot. V. 170 (1861); Schulz. l.c. 42

Fig. 46. Uncommon.

Seed—1.8 × 1.8 × 1.6 mm.; viewed from above round, from the side an elongated oval; irregular; net pentagonal-meshed and medium deep.

Cotyledons—1.1 cm. × 7.9 mm.; turtle green; medium veins marked for short distances; indentation deep and narrow.

Seedling—tall; bright green stem; purple nodes; internode.

Seedling leaves—glabrous or very sparsely setose; shiny; wrinkled; obovate with acuminate base; dentately serrate.

11. **Brassica napobrassica** (L.) Mill. Gard. Dict. ed. 8 (1768) Bailey l.c. 237
B. Napus var. *napobrassica* (L.) Reichb. Mossler's Handb. Gew. 3. ed. II. 1220 (1833)

Swede turnip or rutabaga. Figs. 47 *d*, 48. Cultivated for food. Each of the many strains of Swede turnip according to size and color of seed, or to seedling size, edge of leaf, and intensity of blue glaucous color may be arranged in a series from those that resemble the rapes on the one hand to those close to the turnip on the other. Yellow Williamsburger, Improved Lord Derby, and Stirling Castle are more rapelike, while Improved Lothian and Ewing's Invictus appear to resemble certain turnip strains.

a. Yellow fleshed strains.

Seed—1.76 × 1.72 × 1.68 mm.; with a range of 1.7 to 1.96 mm. dusky with blackish-brown, auburn, deep and dark neutral gray with traces of red brown; more or less cubical; raphe outside of the oval hilum; radicle ridge present; micropyle broad and black.

Cotyledon—Rinneman's green; very slightly glaucous; thicker and more translucent than turnip; veins visible; size and shape variable; two cotyledons are equal and symmetrical. Lycopinoids in cotyledons of seven-day seedling distinguishes it; Tables I and II.

Seedling—strong; quick growing; *no internode*; hairs on inner face of petiole.

Seedling leaves—becoming more glaucous with age; more or less setose; dull; wrinkled to smooth; broadly oval to round; *shallowly* and sharply triserrate.

b. White fleshed Swede turnip or rutabaga.

Uncommon in Canada.

Seed and seedling characters similar to yellow-fleshed strains with the exception that no color tests apply. Exceptions to the presence of hairs on the inner side of the petiole, sometimes used as a distinctive feature of the Swede turnip, are to be found in both turnips and Swedes. The presence of an internode (Gorman and Lafferty, 1931 (18); Robson, 1934 (53)) was also noted in some Swede turnip seedlings. Thus these points are only indicative and the only positive point of distinction between yellow-fleshed Swedes and dwarf Essex rape in the seven-day seedling stage lies in the distinctive color of the young seedlings grown in the dark. Identification of white-fleshed Swede turnips has to be based on the well developed seedling.

12. **Brassica Rapa** L. Spec. Pl. 666, Figs. 124 and 125 (1753)

B. campestris L. var. *Rapa* (L.) Babington, Man. Brit. Bot. ed. 3. 24 (1843)

Tuberous rooted forms.

A. *Brassica Rapa* L. var. *rapifera* Metz. Syst. Beschr. cult. Kohlart. 50 (1833)

B. Rapa L. var. *Rapa* Thellung, Hegi. l.c. 261

B. Rapa L. var. *lorifolia* Bailey, l.c. 240

I. Common turnip. Fig. 49. Our markets offer two classes of common turnip, the one a yellow-, cream- or white-fleshed type of medium or large size and commonly used both for the table and for cattle feed, and the other a white, soft-fleshed, small type used almost exclusively for table use, sometimes referred to as the garden turnip and appearing early in the fall. The former closely resembles the Swede turnip but may be distinguished from it by the fact that its bulb is an enlarged root and hypocotyl whereas the Swede turnip contains also a portion of the enlarged stem. This difference may be illustrated by cutting the full grown schizocorm in halves vertically and noting the point where the root joins the stem.

Seed—1.72 × 1.56 × 1.56 mm. reaching up to 1.84 mm.; *seed coat distinctive*, Figs. 7, 8; Sanford's brown and auburn, fuscous, fuscous black and deep mouse gray; viewed from above oval to round slightly flattened on radicle side; from the side round or oval with the hilum often projecting well above the seed; raphe overlapping the raised, round, medium small, black hilum; slight depression between micropyle and hilum; micropyle long, narrow, and brown.

Cotyledons—9.4 × 4.76 mm.; deep, full, yellow-green later becoming chromium green; dull; thin; smaller in the fall turnip.

Seedling—slender and strong; Tables I and II.

Seedling leaves—thin, develop early; Scheele's green; shiny; soft; more or less wrinkled with sunken anastomosing veins that lose themselves in lateral veins only toward the tip while in annual rape the larger portion of the central vein stops abruptly near to the petiole; setose with soft hairs on lamina often bent to point toward the tip as if blown by a wind; oval; irregularly triserrate.

II. Japanese foliage turnip, shogoin, tennoje. Figs. 50 to 54 *a*. Leaves sold as greens in spring, bulb forming in the fall.

Seed—2.16 × 1.84 × 1.6 mm.; quaker drab, deep and dark quaker drab; testaceous, cocoa and walnut brown; vinaceous drab, deep and dark vinaceous drab, and grayish-brown; *celled outer subepidermal row* in radial section of seed coat; distinguished from *B. pekinensis*, which it resembles, by the large round raphe, by the general shape, and by the view from above, which shows a clear-cut isosceles triangle; primary net shallow in regular rows.

Cotyledon—1.59 × 1.1 cm.; apple green; bright; medium thick; angular.

Seedling—sturdy; tall; primary leaf rapidly elongating; slightly thickened.

Seedling leaves—glabrous or sparsely setose on edges; forest green becoming lighter and brighter; deep veins, the central noticeably broadened; spatulate with lamina forming a ribbon down petiole; coarsely, evenly, and shallowly serrate.

III. Strap leaf turnip. Fig. 51.

Seed—1.68 × 1.6 × 1.48 mm.; deep and dark purplish-gray, Hay's russet and liver brown, and a few seeds of dark purplish-drab, burnt umber and chocolate brown; covered with gray mucilage in contrast to pak toy, which it resembles; viewed from above rectangular, raphe almost outside the small, round black hilum, from the side elongated from top to bottom and flattened and curved on the lower left corner; seed coat resembling that of common turnip.

Cotyledon—6.35 × 7.9 mm.; light bice green; dull, medium thick; central vein prominent; indentation shallow and angular.

Seedling—fine; small; slender; red tinge to stem and petiole.

Seedling leaves—light bice green later becoming Rinneman's green; shiny; wrinkled; veins white, deeply sunken; not glaucous; bristly setose with very short hairs; broadly oval with truncate tip; evenly biserrate, resembling that of Swede turnip.

B. *Brassica Rapa* L. var. *septiceps* Bailey, l.c. 240

Seven-top turnip. Fig. 52. Tender sprouts are sold in the spring but later in season a bulb forms.

Seed—1.6 × 1.4 × 1.2 mm.; neutral gray, deep neutral gray, deep brownish-and dusky drab, dark gray, sorghum and Hay's brown; viewed from above wedge-shaped, distinct radicle ridge, from the side slightly flattened at top of upper right corner and considerably flattened at lower left; hilum very small, oval, white tissues and mucilage extending across it tend, in appearance, to cut seed in half; primary net shallow.

Cotyledon—4.76 × 7.9 mm. but on opening only about 2 × 1 mm. in size; turtle green; dull; medium thick; central vein prominent; heart-shaped; indentation shallow and angular.

Seedling—fine; small; stem white.

Seedling leaves—Schafe's green; rugose; thick; leathery; setulose with bristly hairs; veins deeply cut; slightly more elongated ovate than common turnip; regularly shallowly serrate.

C. *Brassica Rapa* L. var. *periviridis* Bailey, l.c. 243

B. campestris L. var. *chinensis* (L.) Kondo, *Brass.* Spec. in Japan (1917)

Tendergreen or mustard spinach, komatsu-na. Fig. 54 *c.* This plant has been placed in this group on account of its slightly thickened pure white tap root but in Eastern Asia it is usually included, with the other "sprouts" or "na's" in *B. campestris* L. Perhaps that is the better placing. The plant itself is used for greens, while in China in the fall the roots are cut in 1.8 cm. cubes, tied in bundles of six or eight and pickled. In this form they were imported into this country and offered for sale on our markets.

Seed—1.8 × 1.72 × 1.48 mm.; dark vinaceous and dark grayish-brown with a few quaker drab seeds; viewed from above an isosceles triangle slightly rounded at the apex and with the micropyle at the center of the base more as in the common turnip than as in Chinese cabbage, from the side triangular with the right side perpendicular, the lower left corner cut away and the top and other side but slightly rounded; primary net varying; secondary net just visible.

Cotyledon—1.27 cm. × 9.5 mm.; forest green; bright; central vein distinct; indentation deep and curved.

Seedling—low; sturdy; *lumiere green stems.*

Seedling leaves—forest green or lighter in color; glazed; sunken lateral veins; may or may not have a cluster of setose hairs at the slightly enlarged tip of serrations and scattered hairs on edges; broadly oval to round, very slightly acuminate; exceptionally shallowly serrate, almost entire.

13. **Brassica chinensis** (L.) Bailey, l.c. 253

B. campestris L. subsp. *chinensis* L. var. *brassicata* (L.) Burkill, l.c. 1

B. Napus L. var. *chinensis* (L.) Schulz, l.c. 45

Pak choi, white celery mustard. Fig. 54 *b*. This plant in the green state is commonly sold in Canada by Chinese merchants for use as vegetable. It somewhat resembles Swiss chard in its broadened white veins and petioles, which, forming a loose head at the base, lose themselves by dividing and projecting fanwise far into the lamina. This plant has been placed in *Brassica chinensis* L. but possibly it might better have have been included in *B. campestris* L.

Seed—1.6 × 1.28 × 1.48 mm.; deep and dark neutral gray, dark vinaceous, seal brown, and dark grayish-brown; viewed from above rounded, sometimes oval, with a *small raphe* that differs from the large one of *B. pekinensis* Rupr.; *hilum small round and black* like that of common turnip, from the side rounded except for the slight flattening on the lower right side and the decided one on the upper right corner; primary net fine, shallow, and in rows; only slight trace of vinaceous mucilage; micropyle long and narrow.

Cotyledons—7.9 × 4.76 mm.; rivage green; dull; thin; central vein prominent; indentation broad, shallow, rounded in one, narrower, more triangular in the other.

Seedling—small; white succulent stem tinted red.

Seedling leaves—smooth, glabrous, thick, evenly serrate with a tiny sawlike hair at the point of each serration; obovate acuminate; lamina ribbon down stem for short distance.

14. **Brassica campestris** L. Spec. pl. ed. I, II (1753)

B. Rapa L. var. *campestris* (L.) Koch, Syn. Fl. Germ. et Helv. 1, ed. 55 (1835)

A. *Brassica campestris* L. Sp. pl. 666, Fig. 128, 130 (1753); Bailey l.c. 245

B. Rapa L. var. *sylvestris* Lam. f. *campestris* (L.) Koch; Hegi l.c. 259

Wild campestris, field mustard. Figs. 53 to 56 *a*, Figs. 11, 12. Seed gathered from plants growing wild in eastern Canada was identical with that of a sample of bird feed seed cultivated in Argentine and imported into Canada except that raised micropyle was not pronounced.

Seed—1.48 × 1.4 × 1.36 mm.; dull; deep, dark and dusky purplish-gray with some seal brown and dark purple drab; viewed from above almost a square with rounded corners and radicle ridge very slightly visible; from the side, almost round; raphe, with vascular bundle scar appearing as a small white dot, is on well defined oval ring of the brownish-black hilum; micropyle on tip of *extended radicle ridge* causes a depression between it and the raphe not found in the smaller seeded strains of turnip; primary net, distinct with fine mesh, is arranged in circles around raised micropyle.

Cotyledon—8.5 × 4.76 mm.; medium green; indentation narrow.

Seedling—small; first two leaves appearing simultaneously.

Seedling leaves—small; dull; thick; sparsely setulose on upper lamina, under veins, and petioles; elliptical to round often narrowing toward the tip; small, serrated end lobe; primary often with three serrations on one side and four on other; secondary with four biserrations; later leaves becoming cut.

B. *Brassica campestris* L. f. *biennis* Reichb. Deutschl. Fl. I. t. 92 Fig. 4434 B. 106 (1837–38); Syn. Hegi. 261

B. campestris L. *autumnalis* DC. Syn. Schulz l.c. 49

B. Rapa L. var. *sylvestris* Lam. f. *biennis* (Metz.) Alef. Hegi. l.c. 261

B. Rapa L. *oleifera biennis* Metz. Syst. Beschr. cult. Kohlart 50 (1833)

I. Roller rape, oil rape. Fig. 55. Imported as bird feed. Prize roller canary and other bird fanciers demand this seed when possible claiming they can distinguish it by its sweet, mellow, nutty flavor, and by the way birds enjoy it.

Seed—1.7 × 1.7 × 1.55 mm.; aniline black, Sanford's brown, deep and dark livid brown, Morocco red, dark and dusky purplish-gray; viewed from above circular with conspicuous round black hilum and small raphe partly within it; from the side, all corners but lower left are angular and seed is often elongated; seed coat *almost smooth, bright*; primary net shallow, small, round-meshed; hilum, biennial with its cells distinct from those of seed coat and with two strands of mucilage from raphe circling its central scar. Figs. 13, 14.

Cotyledons—7.9 × 6.35 mm.; Rinneman's green; dull; medium thin with prominent midvein; angled near the base of lamina; indentation narrow and arcuate.

Seedling—strong; no internode; leaves whorled; Table I.

Seedling leaves—large; dull; not glaucous; rugose; hirsute above, below, on edges and on petiole; primary elliptical; unsymmetrical; lamina at base acuminate on one side, on other broadly cut at an angle with petiole to form chin of a caricature of a human profile; broadest in center; four or six uneven coarse serrations; later leaves becoming lyrate, with three small segments and three lobes in main section.

C. *Brassica campestris* L. f. *annua* Reichb. l.c. Fig. 4434 *a*; Syn. Hegi. 260

B. Rapa L. var. *sylvestris* Lam. f. *annua* (Metz.) Alef.; Hegi. l.c. 260

B. Rapa L. *oleifera annua* Metz. Syst. Des. Kohl. 51 (1833)

I. Bird rape, red summer rape, Fig. 27. Low grade bird feed. Seeds of wild Brassicas as well as cultivated, of low vitality, are commonly present. Extreme forms grow quickly, branch readily, mature early, and produce an enormous number of seeds. Indeed some produce seed in the garden at a height of four inches. This may be the plant described by Muravieva (1928 (41)) as *B. campestris*, field cabbage.

Seed—2.1 × 1.9 × 1.7 mm.; dull; Sanford's brown, dark livid brown and dusky drab, Hay's brown, russet and liver brown; viewed from above cubical with rounded corners, flattened, slightly less on radicle side; from the side, oval, lower left corner cut away so that seed rests naturally there; primary net, large-meshed, exceedingly shallow, with straight, flat, and broad walls.

Cotyledon—1.43 × 1.27 cm.; medium green, dull, thick, indentation arcuate twice as wide as deep.

Seedling—elongating rapidly; may flower after third leaf.

Seedling leaves—very shiny; forest green; usually glabrous sometimes sparsely setose on under veins or upper lamina; oblong; acute angled at base; regularly serrate.

In the case of all annual rapes if primary or secondary leaf shows an unusual *five*-lobed or serrated *juncea* character, pure seeds of *B. juncea* (L.) Coss. can be expected to segregate out. Closely related strains produce larger plants. Their seeds are smaller, with a deeper-walled small-meshed net and seedling leaves are duller, broader, and softer in texture.

II. Rubsen rape, small red rape. Fig. 58. A strain much used in bird feed. Easily identified by seed.

Seed—1.9 × 1.8 × 1.4 mm., deep livid brown, Hay's brown, warm blackish-brown, deep brownish-drab. Viewed from above ovate to wedge-shaped, always flattened at hilum, from the side, flattened to straight line on right, with top curved slightly upward and left side and base forming a sweeping curve; net fine and irregular, seed coat reticulated. Figs. 9, 10.

Cotyledon—1 cm. × 7 mm.; other slightly smaller; dull, thick; chromium green; midvein distinct; indentation angular.

Seedling—internode; forest green stem tinted red.

Seedling leaves—soft; slightly wrinkled; Rinneman's green; glabrous; broadly elliptical with four irregular serrations.

III. Sweet bird rape. Fig. 59. Imported as bird feed, and rarely for oil. Wide variation.

Seed—1.8 × 1.6 × 1.5 mm.; Hay's russet, Morocco red, liver, dark vinaceous, seal, dusky, and blackish browns. Viewed from above, elliptical with an annual hilum, slightly darker than seed coat, raised in line with curved top of seed; micropyle brown; from the side, elliptical to shield-shaped with upper right top flattened; coat with reticulations elongated lengthwise, secondary net, clear, of fine round mesh. Netting often deeper, larger, and more pronounced.

Seedling—tall, strong, short internode.

Seedling leaves—smooth, setulose above, below, and on petioles; elliptical; truncate tip; each side cleft into four varying, irregular, unsymmetrical lobes or serrations; sometimes erose.

15. **Brassica elongata** Ehrh. Beitrage Naturh. VII. 159 (1792)

Seed—1 × 0.75 × 0.66 mm.; olive gray with very distinctive strip of Natal brown covering the radicle ridge, the tip of which projects above the seed. When seed is crushed and heated with chloral hydrate, the outer kernel skin colors red but the seed coat, although it closely resembles in texture that of *B. arvensis* (L.) Rabenh., does not bleed.

16. **Brassica juncea** (L.) Coss., Bull. Soc. Bot. Fr. VI. 609 (1859)

Salad plants grown in gardens and some oil seeds produced.

Seeds of the species were well defined and had a definite heavy netting. *B. juncea* represents an exceedingly large group. Seeds from different sources were grown and the resultant plants showed astonishing differences, varying in height from a few inches to several feet and otherwise varying from small tender plants to very large and coarse rushlike ones, which might or might not be leafy and branching. The salad forms are the curled and broad leaved varieties plain or savoy type. Seed distinction within the species is difficult.

A. *B. juncea* (L.) Coss., wild form

Brown mustard, wild mustard. Fig. 63. In several samples of grain screenings from Western Canada this wild *juncea* seed constituted somewhat less than 1% of the *small* seeds. The percentage is much smaller in Ontario screenings.

Seed—1.6 × 1.52 × 1.4 mm.; dark Natal brown and bone brown; often perfectly round but sometimes elongated, net shallow and superficial. Distinguished by the very tall-celled outer epidermal row that appears when radial section of seed coat is immersed in water. Fig. 15.

Cotyledons—1.56 × 1.27 cm.; Rinneman's green; slightly American green glaucous; dull; medium thick; edges and prominent central vein tinted red; cordate with distinct angle at base of side; indentation deep, narrow, and curved.

Seedling—sturdy; green stem with bronze tint; *no marked tendency to whorl*; Table I.

Seedling leaves—large; shiny; smooth; *very dark green*; veins sunken; sparsely setose on veins, edges, and petiole; very rarely symmetrical; fragments of lamina usually but not always appearing down the stem; in other cases base of the lamina parted; oblong with slight truncate tip.

B. Leaf mustards

I. *B. juncea* var. *crispifolia* Bailey, l.c. 41. (1922)

Curled mustards. Fig. 61. Common strains; curled mustard, giant southern curled, and ostrich plume.

Seed—1.44 × 1.4 × 1.36 mm.; vinaceous and dark vinaceous brown, deep brownish- and dusky drab and a few deep and dark mouse gray; viewed from above circular, from the side egg-shaped; net comparatively *shallow* and of *medium-sized mesh.* Radial section in water shows medium tall-celled outer epidermal row.

Cotyledons—1.27 cm. × 9.5 mm.; *ambrical green*; glaucous; dull; thick; slight angle at base giving a more rectangular appearance than that of the average heart-shaped *juncea* cotyledon; central vein only; indentation irregular.

Seedling—medium fine; greenish-white stems.

Seedling leaves—characteristically *shiny*; sparsely setose with short hairs on edges of primary leaf and on lamina in secondary leaf; some much more elaborately cleft or parted than others.

II. *B. juncea* var. *foliosa* Bailey, l.c. 263 (1930)

Common strains—Florida broad leaf, elephant's ear, chirimen savoy leaf, new Chinese and Chinese smoothleaf mustards, Fordhook fancy. Fig. 16.

a. Florida broadleaf; smooth and Savoy types, some cut and curled.

Seed—1.56 × 1.48 × 1.4 mm.; mixture of vinaceous and dark vinaceous brown, deep brownish- and dusky drab, buff and olive browns; viewed from above rectangular, from the side showing an elongation on the upper left side topped by a raised micropyle; net large with deep perpendicular walls. Section of seed coat mounted in water showed only a trace of shallow cells in outer epidermis.

Cotyledons—9.5 × 4.7 mm.; Rinneman's green; glaucous American green tint; translucent; flattened heart-shaped with lamina ribbon down stem.

Seedling—medium; white stem with red tint or green stem with bronze tint; petiole grooved. Fig. 62.

Seedling leaves—setose; shiny; lighter green; veins white; pronounced; elongated, oblanceolate, ribbon lamina down petiole for a short distance; irregularly, coarsely and shallowly serrate.

b. Guy toy. Fig. 60.

These plants as greens may be purchased from Chinese merchants and are recommended because of their more pungent flavor.

Seed—1.32 × 1.28 × 1.2 mm.; mixture of brownish- and dusky drab; liver, Hay's, and chocolate brown; net small-meshed.

Seedling—considerable variation.

c. Taka-na, a Japanese mustard.

Seed—1.5 × 1.3 × 1.3 mm. in size, and haematite red, Prussian red, dark vinaceous drab, and dark grayish-brown in color.

C. Oil producing mustards. Brown mustards.

Samples of seed were received from European universities. The similarity of the seeds of different strains gave little clue to the variations in the mature plants, some of which, growing much branched and rankly to a height of 4 ft., seemed to justify the name "*juncea*", which is derived from the latin "junceus" meaning "rushlike". Schulz (l.c. 55) points out two varieties of *juncea* seeds, variety *lutea* Batalin, honey yellow, and variety *fusca* Batalin, darker in color. In the lighter colored oil group the seeds of *fusca* are Hay's russet, liver brown, with a few dusky drab and vinaceous brown. The oil seeds usually have a thick-walled heavy net of large mesh through which is seen the alveolate seed coat. Radial sections of the seed coat show an *uncelled thick subepidermal row.*

It is in this group that Burkill places *B. Besseriana* Andrz. with its large oval dark brown seeds and also the "Rai" of India and *S. ramosa* Roxb.

17. **Brassica integrifolia** (West) Schulz, ap. urb. Symb. antill. III. 3. 509 (1901)

Fig. 64. Not common.

Seed—1.2 × 1.2 × 1.5 mm.; honey yellow; brown hilum; raised white raphe; small brown micropyle; primary and secondary nets straight-sided, distinct.

Cotyledons—1.12 cm. × 8 mm.; dark blue green; hypocotyl red; petiole elongated; cotyledon heart-shaped with flattened sides; indentation shallow, narrow, and arcuate.

Seedling—tall; *strong*; *coarse*; stem green with purple tint.

Seedling leaves—dark with purple tint; whorled; wrinkled; net-veined; setulose above and below; obovate; broadly and unevenly dentate; fragments of lamina down petiole.

18. **Brassica nigra** (L.) Koch, in Rühling's Deutschl. Fl. ed. 3, IV. 713 (1833)

Black mustard. Fig. 66. Seeds of black *B. arvensis* type are sometimes incorrectly called black mustard. These are not included in this paper. Six varieties of *B. nigra* are listed by Schulz (1919) but are not grown to any extent here, nor are the seeds of *B. nigra* a common impurity in our *Brassica* seeds.

Seed—1.58 × 1.3 × 1.2 mm.; ranging from 1.44 to 1.6 mm.; Fig. 15; color varying according to variety from dark red to the buff brown of the *juncea*; viewed from above circular, flattened at micropyle, from the side irregular, elongated, and often simulating other *cruciferous* seeds more than *Brassica*; net deep-meshed covering the seed and if removed would leave the seed coat smooth and shiny.

Cotyledon—6.35 × 8 mm.; deep dull yellow-green with the typical tint of American green; thick; smooth; heart-shaped; indentation arcuate.

Seedling—fine; small; decided internode; stem glabrous.

Seedling leaves—deep; dull, yellow-green; glabrous or more or less setose; waxy; straight sunken veins; unsymmetrical; crenately and alternately lobed; oblanceolate to oval.

19. **Brassica arvensis** (L.) Rabenh. Fl. lusatica I. 184 (1839)

B. sinapistrum Boiss. voy. Bot. Midi de l'esp. II. 39 (1839–1845)

Wild mustard. Fig. 65. Of the many varieties of *arvensis* only two growing in our fields and cultivated crops are well known. Fernald has reported finding in eastern Canada another variety, *Schbuhriana* Reichb., which is distinguished by a slender silique 1.5 to 2 mm. thick. No sample of seed of this variety was available.

a. Seed—deep dark dusky gray and Prussian red; coat rougher than Type *b*; spherical, 1.6 × 1.6 × 1.6 mm.

b. Seed—small; smooth; spherical; russet vinaceous, sorghum brown, Hay's brown, light seal brown; mature portion of the seed coat, if boiled in a solution of chloral hydrate, bleeds, i.e. traces of *red liquid* appear to flow very slowly from the reddened cell; Prussian red seeds of first variety are not sufficiently mature to test until a portion of coat darkens.

Cotyledons—9.5 × 6.35 mm.; medium green; smooth; light glaucous coating of blue when young; larger in black-seeded variety; may or may not show red color; indentation deeply curved.

Seedling—slow growing; sturdy; soon becoming ragged and weedy; stems greenish-white with purple tint; unable to separate two varieties by seedling. Table I.

Seedling leaves—smooth; decidedly pubescent with short hairs above, below, on the edges and petioles; thin, elongated linear to lanceolate; upper third of the leaf narrowing in a series of two or three right-angled steps to the truncate tip; often narrows toward the petiole in the same manner; irregularly entire or crenately repand; biserrate; symmetrical.

20. **Brassica alba** Rabenh. Fl. Lusatica I. 185 (1839)
Sinapis alba L.

White mustard; imported for the manufacture of table mustard. Fig. 67.

Seed—2.25 × 2.25 × 2 cm. with range from 1.5 to 2 cm.; large; honey yellow; porous coat; raphe pronounced from side view; distinguished by cross section of seed coat or by immersing half of the seed in water and marking the behavior of mucilage.

Cotyledons—1.59 cm. × 7.9 mm.; leaf green; dull; thick; central vein and *two pairs of lateral veins* very deeply sunken; indentation broad, semicircular but irregular; petiole pubescent.

Seedling—large; medium fine; pubescent and touched with bronze or purple. Table I.

Seedling leaves—pubescent on petioles, under veins and edges; pinnately lobed.

21. **Brassica dissecta** Boiss. l.c. 40; Schulz syn. 132

Seed—brown; from side view raphe less prominent, otherwise appearing as *B. alba*.

Cotyledons—1.8 × 1.1 cm.; pronounced veins; indentations broad, semicircular; setose on petiole.

Seedlings—similar to *B. alba*.

Seedling leaves—similar to those of *B. alba*, but very coarsely lobed.

22. **Brassica barrelieri** (L.) Janka. in Termesz. Fig. VI. 179 (1882)

B. subularia Pers. Syn. II, 207 (1807); Schulz Syn. 65

Seed—0.75 mm. in diameter, perfectly round except for the slightly small round hilum capped with mucilage; dark red brown color with a net of hexagonal mesh so large that only 9 or 10 meshes reach across the seed.

KEY FOR DISTINCTION OF *Brassica* SEEDS BY MEANS OF THE SEED AND OF SEED COAT STRUCTURE

For placement of seed:

1. Viewed from above with micropyle, raphe, and hilum visible.
2. Viewed from side with radicle at left and raphe above the micropyle and the hilum.

NOTE: Brassica seeds are identified only by the intimate knowledge and the segregation of a multiplicity of detail.

a. Seeds honey yellow in color.

b. Seed coat heavily netted, under 1·3 mm. dia.---17. *B. integrifolia* (West) Schulz.

c. Seed over 1·3 mm. dia.---16. *B. juncea* (L.) Coss.

b. Seed coat porous; glaucous; seed dia. over 2 mm.---20. *B. alba* (L.) Rabenh.

b. Seed coat porous; not glaucous; seed under 2 mm. dia.; oleraceous radicle and shape---3B. *B. oleracea* L. var. *acephala* DC.

b. Seed coat reticulated---14. *B. campestris* L.

a. Seeds darker than honey yellow.

d. Seed coat porous; glaucous; dull cinnamon brown; over 2 mm. dia.---21. *B. dissecta* Boiss.

d. Seed coat smooth; palisade cells tall, narrow, tightly packed, and of uniform height.

e. *Palisade* cells of seed coat *not* bleeding in chloral hydrate; seed spherical, approx. 1 mm. dia.; dull; strip of brown color covering the radicle---15. *B. elongata* Ehrh.

e. Palisade cells of seed coat bleeding in chloral hydrate; seed spherical or elongated; dull dark dusky gray or shiny purplish-gray (two varieties)---19. *B. arvensis* (L.) Rabenh.

d. Seed coat prominently netted.

f. Seed usually elongated; primary net in seed coat with slanting walls, seed coat below net, shiny, smooth; from side, seed falls away beneath the micropyle to form a perpendicular radicle ridge---18. *B. nigra* (L.) Koch.

f. Seed tending to spherical; raphe raised; primary net on seed coat with narrow straight-sided walls usually mucilage-tipped; seed coat below dull, alveolate; from side, seed beneath micropyle full and rounded---16. *B. juncea* (L.) Coss.

g. Subepidermal layer of radial section crushed but in water an outer row of shallow cells appears---16B. *B. juncea* (L.) Coss.

g. Subepidermal layer in water shows an outer row of deep cells---16A. *B. juncea* (L.) Coss.

d. Seed coat neither smooth, porous, or prominently netted.

h. Seeds vinaceous, glaucous, from above, smooth, wedge-shaped with rounded sides; from side, seed slants from the *large* raphe forming an elbow near the middle or in the upper half of the right side.

i. Seeds over 1·5 mm. in breadth; reticulations very fine, shallow, round, irregular; crushed subepidermal layer; special bodies in the pigment layer---9. *B. pekinensis* Rupr.

i. Seeds over 1·5 mm. in breadth; large raphe angling over the hilum from a depression; reticulations fine, irregular; outer row of subepidermal layer is celled---12A. II. *B. Rapa* L. var. *lorifolia* Bailey

i. Seeds over 1·5 mm. in breadth; from above, large raphe touching the micropyle surrounded by white mucilage; from the side, elbow forming upper right corner; reticulations deep, in rows ..... 12C. *B. Rapa* L. var. *periviridis* Bailey

i. Seeds 1·5 mm. in breadth or under; reticulations fine, tending to regular rows, micropyle long, narrow, black ..... 13. *B. chinensis* (L.) Bailey

h. Seeds deep dark and blackish mouse gray; 1·9 mm. in breadth; from above, circular; from the side, shield-shaped rounded at top; shallow square-meshed net; spots of gray mucilage ..... 4. *B. alboglabra* Bailey

h. Seed from above, brown or gray oleraceous colors either in coat or in mucilage on coat; rough; large, oval hilum, raphe outside.

j. Radial section of seed coat shows tumbler cells loosely packed, of uniform height, with uniformly thickened walls; outer subepidermal row celled (sheep kale, asparagus kale, and ragged Jack kale excepted) with conical projections of mucilage in center of base of each cell ..... 3. *B. oleracea* L.

NOTE: Varieties of *B. oleracea* distinguished by shape of seeds (Figs. 18, 19) and by color, as indicated in the following portion of the key.

k. Deep mouse gray, spherical.

l. Large ..... 3B. *acephala* DC.
Ragged Jack kale
Asparagus kale

l. Small ..... 3B. *acephala* DC.
Sheep kale

k. Seeds of clay color and tawny olive conspicuously present ..... 3B. *acephala* DC.
Green kale
Variegated kale

k. Seeds of Saccardo's umber and sepia, mouse gray, brownish-drab with the occasionally warm sepia, russet, or hazel; seed coat very thin ..... 3E. *gemmifera* (DC.) Zenk.

k. Seeds of sorghum brown; Hay's brown and light seal brown, shades of brownish-drab, Morocco red, claret brown, and mouse gray ..... 3H. *gongylodes* L.

k. Seeds of drab and hair brown, of Natal and bone brown ..... 3G. *botrytis* L.
cauliflora (Gars.) DC.

k. Seeds of hazel and chestnut brown, of Natal brown and bone brown, few deep olive gray, and wood brown ..... 3G. *botrytis* L.
asparagoides DC.

k. Seeds of dark olive buff, mouse gray, dark olive gray, olive brown, and clove brown ..... 3B. *acephala* DC.
Marrow kale

k. Seeds of grayish olive, olive gray, olive brown, and clove brown ..... 3A. *ramosa* (DC.) Alef.

k. Seeds of vinaceous drab, fuscous and fuscous black, pecan brown, and Rood's brown ..... 3D. *sabauda* L.

k. Seeds of testaceous, cocoa brown, walnut brown and burnt umber, deep brownish drab and dusky drab, and blackish brown, fuscous, and a few dark olive gray ..... 3F. *capitata* L.

h. Seeds uniformly purplish, neutral or mouse gray, dusky or blackish-brown; from above, smooth, rectangularly spherical, flattened in hilum region; raphe outside oval hilum.

j. Radial section of seed coat with cells of subepidermal layer crushed; tumblers with flared brims heavily lignified above and turned back and downwards to form a depression at junction of two brims; lumen of tumbler broader than double wall.

m. Cotyledons grown in dark, large, empire yellow in color ..... 10. *B. Napus* L.

n. Seeds spherical; no anthocyanins in seedlings; radial section with irregularly broad double tumbler walls with marked depression; see seedling ..... 10A. *B. Napus* L. f. *biennis* Thell.

n. Seeds spherical; no anthocyanins in seedlings; radial section with narrower, taller, double tumbler walls and narrower, deeper depressions; see seedling ..... 10B. *B. Napus* L. f. *annua* Thell.

n. Seeds cubical, resting naturally with hilum above; purple and bronze anthocyanins in seedling; see seedling ..... 11. *B. napobrassica* (L.) Mill. White-fleshed Swede turnip

m. Cotyledons grown in dark, Naples yellow or chrome in color ..... 11. *B. napobrassica* (L.) Mill. Yellow-fleshed Swede turnip

h. Seeds from side, a small white raphe angles over the raised round black hilum from the right of which the top slants down to the upper right corner, lower left side flattened; micropyle long, narrow and brown; seed coat characteristic; typical color, fuscous.

j. Radial section of seed with the lignified portion of the inside tumbler wall tapering in a graceful curve to a peak at junction of two tumbler brims; outer subepidermal row crushed but not dipping down into the lumen of the tumbler ..... 12. *B. Rapa* L.

o. Seeds with smallest dia. 1·2 mm.; color typical; from above, wedge-shaped; distinct radicle ridge; straight line of micropyle and tissue over raphe and hilum appears to divide seed ..... 12B. *B. Rapa* L. var. *septiceps* Bailey

o. Seeds over 1·2 mm. dia.; from above, rectangular; confused with *B. chinensis* L. ..... 12A. *B. Rapa* L. var. *rapifera* Metz. Strap leaf turnip

o. Seeds over 1·2 mm. dia.; from above and from side, oval to round ..... 12*A*. *B. Rapa* L. var. *rapifera* Metz. Common turnip

h. Seed colors Hay's brown, russet, or Morocco red, indicated; seed coat reticulations becoming flat-topped and ranging close to netting; from above, shield-shaped, from side, same, sometimes with slight flattening on right top or lower left.

j. Radial section of seed coats, with palisade cells more cylindrical; with the inner wall of the tumbler cells angled where its lignified portion tapers slightly to the brim; outer subepidermal row crushed ..... 14. *B. campestris* L.

p. Seeds approximately 1·7 mm. dia.; bright; regularity of tumbler walls and size of lumen easily noted in simple surface view slide; seed coat with small, medium deep, and irregular primary net, satin finish; indication of Morocco red ..... 14. *B. campestris* L. f. *biennis* Reichb.

p. Seeds almost spherical, 1·4 mm. dia.; sometimes elongated; thin gray glaucous coat; radicle ridge extending above the seed; note surface view slide ..... 14. *B. campestris* L. f. *annua* Reichb.
Wild form

p. Seeds larger, shield-shaped; typical but of wide variation; tendency to russet and Hay's brown; shallow or deep flat-topped reticulations with fine or coarse mesh; see surface view slide ..... 14. *B. campestris* L. f. *annua* Reichb.
Bird rape

KEY TO *Brassica* SEEDLINGS

a. Stem decidedly setose, cotyledons large with two pairs of distinctly prominent veins.

b. Seedling leaves lyrate, lateral lobes unequally crenate ..... 20. *B. alba* (L.) Rabenh.

b. Seedling leaves parted, lateral lobes broadly dentate ..... 21. *B. dissecta* Boiss.

a. Stem glabrous.

c. Cotyledons light green.

d. Stems white.

e. Seedlings glaucous, oleraceous, seedling leaves olivine green ..... 4. *B. alboglabra* Bailey

e. Seedlings not glaucous or oleraceous; stems and veins white and succulent.

f. Cotyledons exceedingly large, acuminate at base, apple green ..... 9. *B. pekinensis* Rupr.

f. Cotyledons small, rivage green ..... 13. *B. chinensis* L.

d. Stems lumiere green.

g. Seedling leaves rapidly becoming oblanceolate, lamina ribbon down stem ..... 12A. *B. Rapa* L. var. *rapifera* Metz.
Shogoin turnip

g. Seedling leaves small, round to oval attenuate at base with short petioles ..... 12C. *B. Rapa* L. var. *perviridis* Bailey

d. Stems apple green—seedling leaves apple green, soon becoming cut ..... 3B. *B. oleracea* L. var. *acephala* DC.
Green kale
3C. Palm tree kale

c. Cotyledons medium green.

h. Cotyledons fleshy, waxed; seedling leaves with greenish glaucous coat.

i. Seedling leaves with the green overshadowed by red, blue, purple, or white ..... 3B. *B. oleracea* L. var. *acephala* DC.
Variegated kale
White ribbon kale

i. Seedling leaves with no anthocyanins visible.
j. Cotyledons when young puffing in center like a balloon top.
k. Stems, petioles, and veins succulent, thick; seedling leaf stiffly erect and almost entire---- 3F. *B. oleracea* L. var. *capitata* L.

k. Stems, petioles, and veins not so succulent; finer; seedling leaf with petiole more gracefully arched, serrately cut---------- 3D. *B. oleracea* L. var. *sabauda* (L.) Martens

j. Cotyledons when young with indentations unsymmetrical, one being broad and almost straightly cut, other arcuate.
l. Cotyledons on opening standing perfectly erect with ventral surfaces together; primary leaf round, entire with short petioles---------- 3E. *B. oleracea* L. var. *gemmifera* (DC.) Zenk.

l. Cotyledons opening not standing perfectly erect; primary leaf with petiole gracefully elongated-- 3H. *B. oleracea* L. var. *gongylodes* L.

j. Cotyledons when young perfectly circular except for the indentation and junction with the petiole-- 3B. *B. oleracea* L. var. *acephala* DC. Marrow kale

j. Cotyledons when young forming a rectangular table.
m. Table with angular pieces cut from each of four sides---------- 3G. *B. oleracea* L. var. *botrytis* L. subvar. *cauliflora* (Gars.) DC.

m. Table with more rounded pieces cut from each of four sides---------- 3G. *B. oleracea* L. var. *botrytis* L. *asparagoides* DC. Broccoli (Walcheren)

j. Cotyledons with none of these characters.
n. Seedling leaves setose---------- 3B. *B. oleracea* L. var. *acephala* DC. Asparagus kale

n. Seedling leaves smooth.
o. Seedling leaf irregular; elliptical; petioles and base of central vein enlarged---------- 3B. *B. oleracea* L. var. *acephala* DC. Collards

o. Seedling leaf irregular; ovate; petioles and base of central vein not enlarged---------- 3G. *B. oleracea* L. var. *botrytis* L. *asparagoides* DC. Sprouting broccoli

o. Seedling leaf irregular; broadly cuneate; veins not enlarged---------- 3A. *B. oleracea* L. var. *ramosa* Alef. Thousand-headed kale

h. Cotyledons neither fleshy nor waxed but leaflike.
p. Cotyledons thin; seedling leaves American green, glaucous.
q. Seedling leaves crenately and irregularly lobed; lyrate----------18. *B. nigra* (L.) Koch.

q. Seedling leaves commonly with five serrations or lobes; often whorled; cotyledons obcordate-----16. *B. juncea* (L.) Coss.

q. Seedling leaves symmetrical, diminishing from center toward each end in a series of irregular, broadly obtuse angles; becoming weedy ..... 19. *B. arvensis* (L.) Rabenh.

p. Cotyledons thin; seedling leaves not glaucous.

r. Cotyledons on opening second smallest of the Brassicas (2 mm. diam.) ..... 12B. *B. Rapa* L. var. *septiceps* Bailey

r. Cotyledons on opening larger; seedling leaves with four serrations or lobes on each side.

s. Seedling leaves with some hairs bent in center to form a right angle ..... 12. *B. Rapa* L.

s. Bent hairs may or may not be present, seedling leaves appearing early wrinkled, one or more dentations or crenations are combined with serrations in characteristic pattern ..... 12. *B. Rapa* L. var. *rapifera* Metz.
Common turnip

s. No bent hairs present; seedling leaves smooth.

t. Seedling leaves shiny, flowering early, decided internode ..... 14C. *B. campestris* L. f. *annua* Reichb.
Large bird rape

t. Two seedling leaves appearing simultaneously; dull; secondary leaf biserrate, later leaves incised ..... 14A. *B. campestris*
Field mustard

t. Seedling leaves unsymmetrical, four irregular lobes on each side ..... 14C. *B. campestris* L. f. *annua* Reichb.
Summer rape

r. Cotyledons on opening larger, seedling leaves with four and six serrations, later leaves elongating and becoming lyrate; decidedly setose ..... 14B. *B. campestris* L. f. *biennis* Reichb.
Roller rape, oil rape

p. Cotyledons thick; tough; translucent; distinct pair of lateral veins; seedling leaves gray glaucous.

u. Seedling leaves only slightly gray glaucous; wrinkled, commonly with six groups of serrations; no internode.

v. Primary seedling leaf broadly oval to round; biserrate; hirsute ..... 11. *B. napobrassica* (L.) Mill.

v. Primary seedling leaf gray glaucous below, bright green above; elliptical with truncate tip; serrations singly or in groups; setose; lamina ribbon down stem ..... 12A. *B. Rapa* L. var. *rapifera* (Metz.) Bailey
Strap leaf turnip

u. Seedling leaves with heavy, gray, glaucous coat; smooth; three or four serrations or lobes; internode; primary seedling leaf with miter-shaped base capped with a prominent dome.

w. Cotyledons large, resembling two circles touching at midrib; primary leaf generally with no segments down stem; elongated dome; broadest in center ..... 10. *B. Napus* L. f. *annua* Thell.
Winter oil rape

w. Cotyledons large, almost as long as wide; primary seedling leaf generally four-lobed; segments down stem; flattened dome; broadest at top of base ..... 10. *B. Napus* L. f. *biennis* Thell.

c. Cotyledons very dark green.
x. Cotyledons kidney-shaped; seedling leaves sharply, unevenly, serrately cut; glabrous; ovate ..... 6. *B. insularis* Moris
x. Cotyledons heart-shaped.
y. Cotyledons large; elongated; seedling leaves with repand edges ..... 7. *B. cretica* Lam.
y. Cotyledons small ..... 2. *B. macrocarpa* Guss.
x. Cotyledon neither kidney- nor heart-shaped.
z. Indentation wide almost straightly cut ..... 1. *B. balearica* Pers.
z. Indentation not so wide, more arcuate.
Z. Seedling leaves glaucous blue above, hoary gray beneath ..... 8. *B. incana* Ten.
Z. Seedling leaves rectangular with gray, glaucous petioles ..... 5. *B. Robertiana* Gay.

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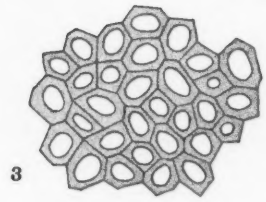
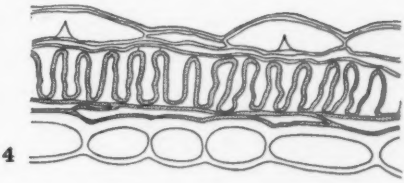
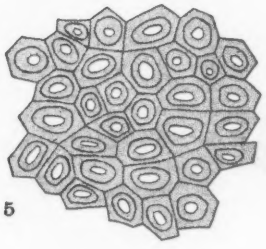
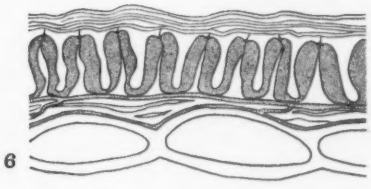
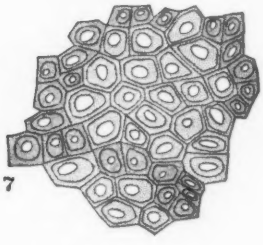
NOTE: *Figs. 3-67 will be found on pp. 579-587.*

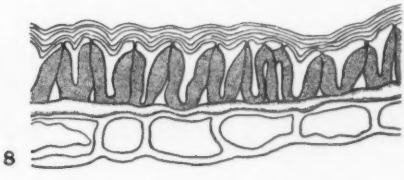


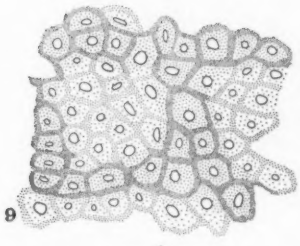


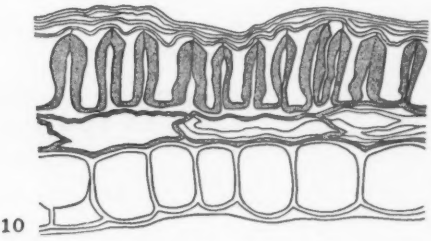












FIGS. 3 *to* 10 *are camera lucida drawings of Brassica seed coats* (×*280*).

FIG. 3. *B. oleracea L. var. gemmifera (DC.) Zenker. Brussels sprouts. Surface view.*

FIG. 4. *B. oleracea L. var. gemmifera (DC.) Zenker. Brussels sprouts. Radial section.*

FIG. 5. *B. Napus L. var. arvensis (Lam.) Thell. f. biennis (Shubl. et Mart.) Thell. Dwarf Essex rape. Surface view.*

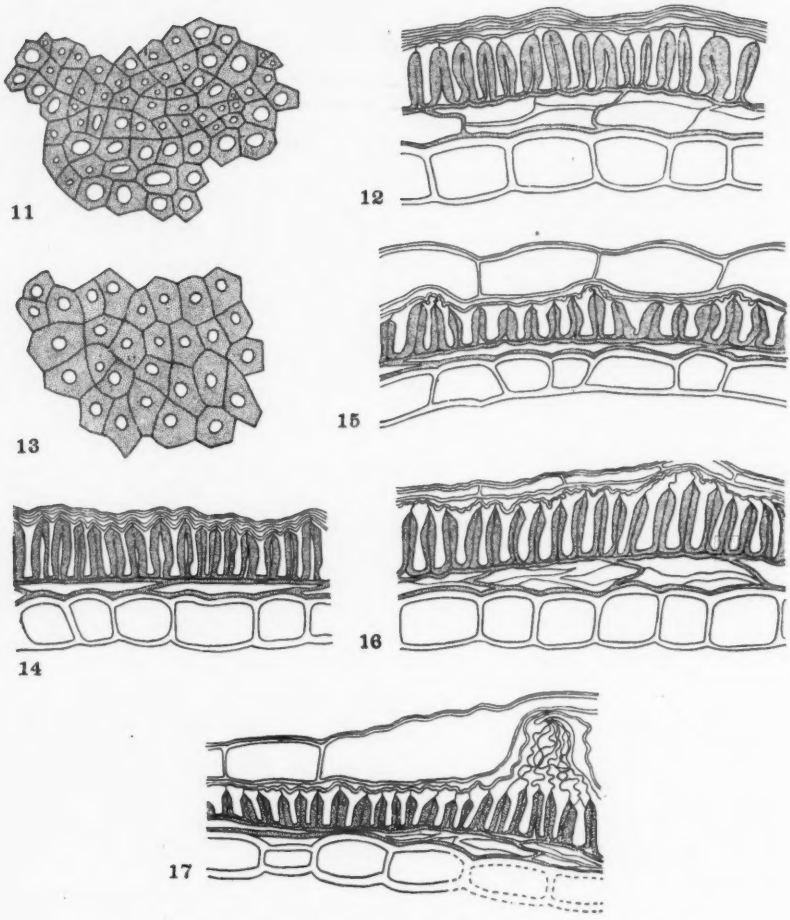
FIG. 6. *B. Napus L. var. arvensis (Lam.) Thell. f. biennis (Shubl. et Mart.) Thell. Dwarf Essex rape. Radial section.*

FIG. 7. *B. Rapa L. var. rapifera Metz. Devonshire turnip. Surface view.*

FIG. 8. *B. Rapa L. var. rapifera Metz. Devonshire turnip. Radial section.*

FIG. 9. *B. campestris L. α annua Reichb. Annual rape. Surface view.*

FIG. 10. *B. campestris L. α annua Reichb. Annual rape. Radial section.*



FIGS. 11 TO 17 *are camera lucida drawings of Brassica seed coats* (×280).

FIG. 11. *B. campestris L., wild form. Radial section.*
FIG. 12. *B. campestris L., wild form. Surface view.*
FIG. 13. *B. campestris L. α biennis Reichb., biennial rape. Surface view.*
FIG. 14. *B. campestris L. α biennis Reichb., biennial rape. Radial section.*
FIG. 15. *B. juncea (L.) Coss., wild form. Radial section.*
FIG. 16. *B. juncea (L.) Coss., smooth leaf form. Radial section.*
FIG. 17. *B. nigra (L.) Koch. Radial section.*

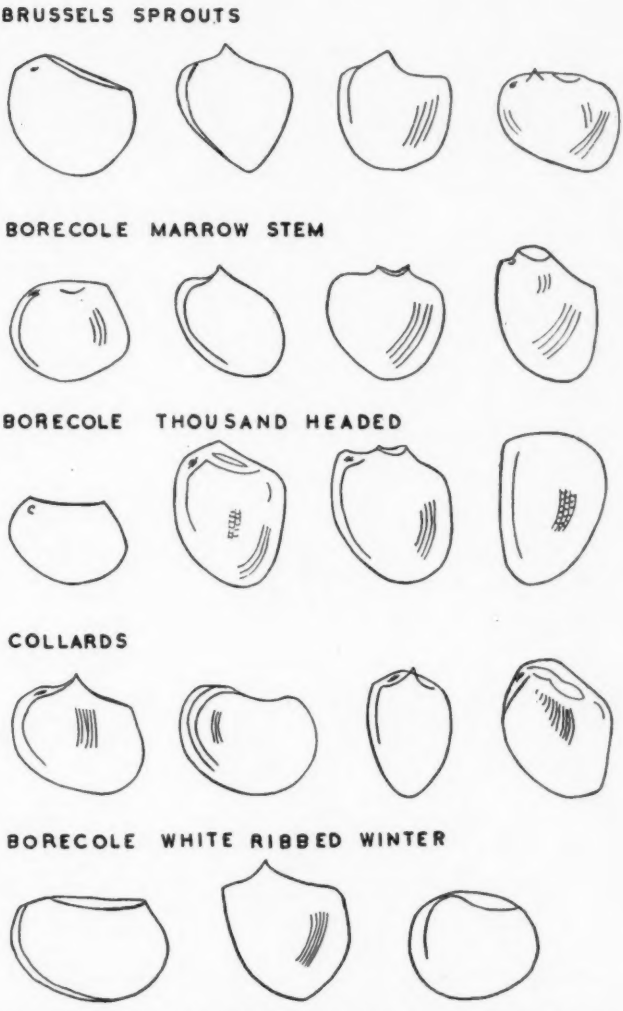


FIG. 18. *Lateral view of the following varieties and strains of Brassica oleracea L., illustrating representative shapes on which distinction of seeds may be based.*

Brussels sprouts. B. oleracea L. var. gemmifera (DC.) Zenker.
Marrow borecole. B. oleracea L. var. acephala DC.
Thousand-headed borecole. B. oleracea L. var. ramosa (DC.) Alefeld.
Collards. B. oleracea L. var. acephala DC.
White-ribbed winter borecole. B. oleracea L. var. acephala DC.

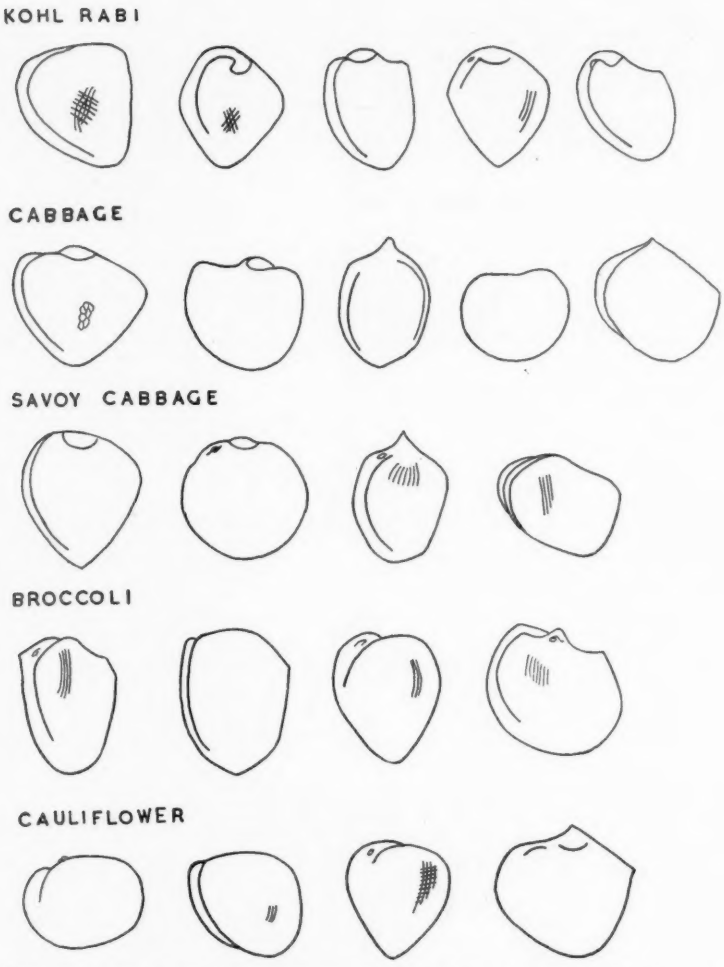
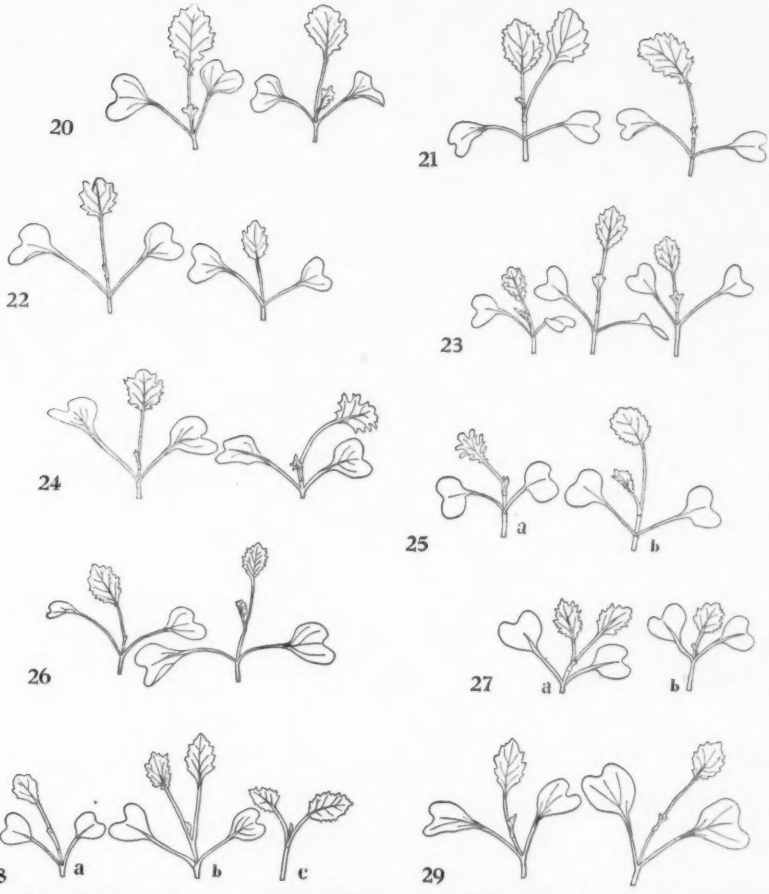


FIG. 19. *Lateral view of the following varieties and strains of Brassica oleracea L. illustrating representative shapes on which distinction of seeds may be based.*

Kohlrabi. B. oleracea L. var. gongylodes L.
Cabbage. B. oleracea L. var. capitata L.
Savoy cabbage. B. oleracea L. var. sabauda (L.) Marten.
Broccoli. B. oleracea L. var. botrytis L. subvar. asparagoides DC.
Cauliflower. B. oleracea L. var. botrytis L. subvar. cauliflora (Gars.) DC.



FIGS. 20 TO 29. *Brassica seedlings illustrating representative characters on which distinction of seedlings may be based. Drawings of photographs of growing plants.*

FIG. 20. *B. balearica Pers.*

FIG. 21. *B. macrocarpa Guss.*

FIG. 22. *B. oleracea L. var. ramosa (DC.) Alefeld. Flanders kale.*

FIG. 23. *B. oleracea L. var. ramosa (DC.) Alefeld. Thousand-headed kale.*

FIG. 24. *B. oleracea L. var. acephala DC. Green kale.*

FIG. 25, *a. B. oleracea L. var. acephala DC. Ragged Jack kale.*
b. B. oleracea L. var. acephala DC. Asparagus kale.

FIG. 26. *B. oleracea L. var. acephala DC. Collards.*

FIG. 27, *a. B. oleracea L. var. acephala DC. White ribbon kale.*
b. B. oleracea L. var. acephala DC. Cottagers kale.

FIG. 28, *a. B. oleracea L. var. bullata DC. subvar. palmifolia DC. Palm tree kale.*
b. B. oleracea L. var. acephala DC. Milan kale.
c. B. oleracea L. var. acephala DC. Variegated kale.

FIG. 29. *B. oleracea L. var. acephala DC. Marrow kale.*

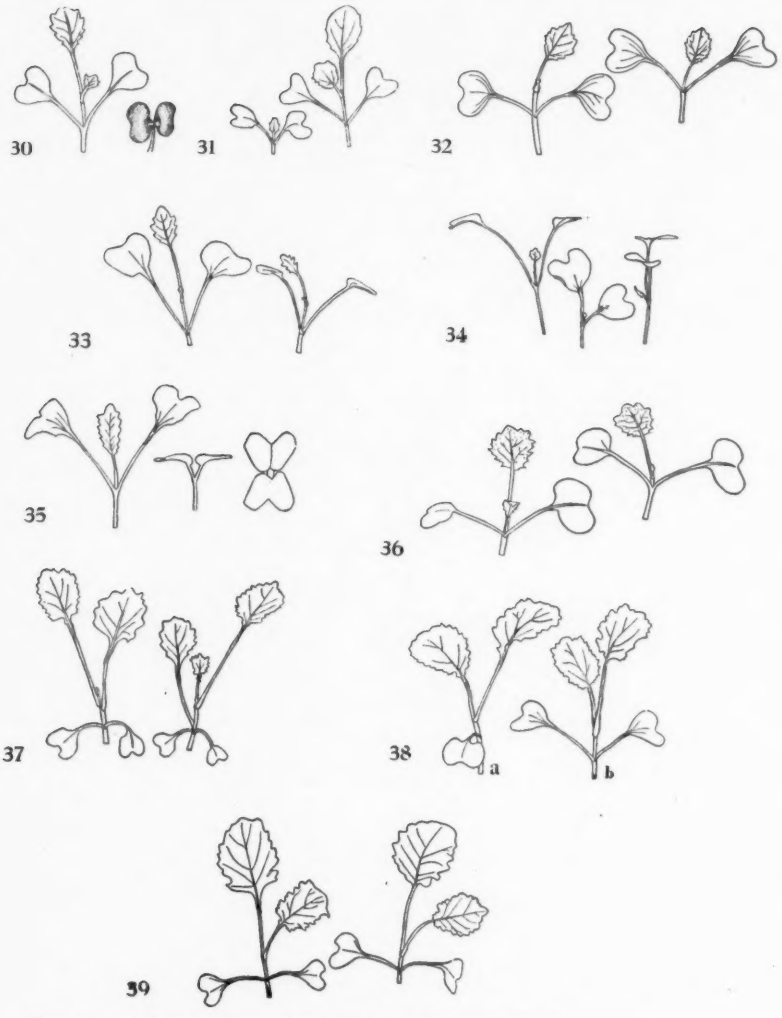


FIG. 30. *B. oleracea L. var. sabauda (L.) Mart. Savoy cabbage.*
FIG. 31. *B. oleracea L. var. gemmifera (DC.) Zenk. Brussels sprouts.*
FIG. 32. *B. oleracea L. var. capitata L. Cabbage.*
FIG. 33. *B. oleracea L. var. gongylodes L. Kohlrabi.*
FIG. 34. *B. oleracea L. var. botrytis L. subvar. asparagoides DC. Sprouting broccoli.*
FIG. 35. *B. oleracea L. var. botrytis L. subvar. cauliflora (Gars.) DC. Cauliflower.*
FIG. 36. *B. alboglabra Bailey. Chinese kale.*
FIG. 37. *B. Robertiana Gay.*
FIG. 38, *a. B. insularis Moris.*
b. B. incana Tenore.
FIG. 39. *B. cretica Lam.*

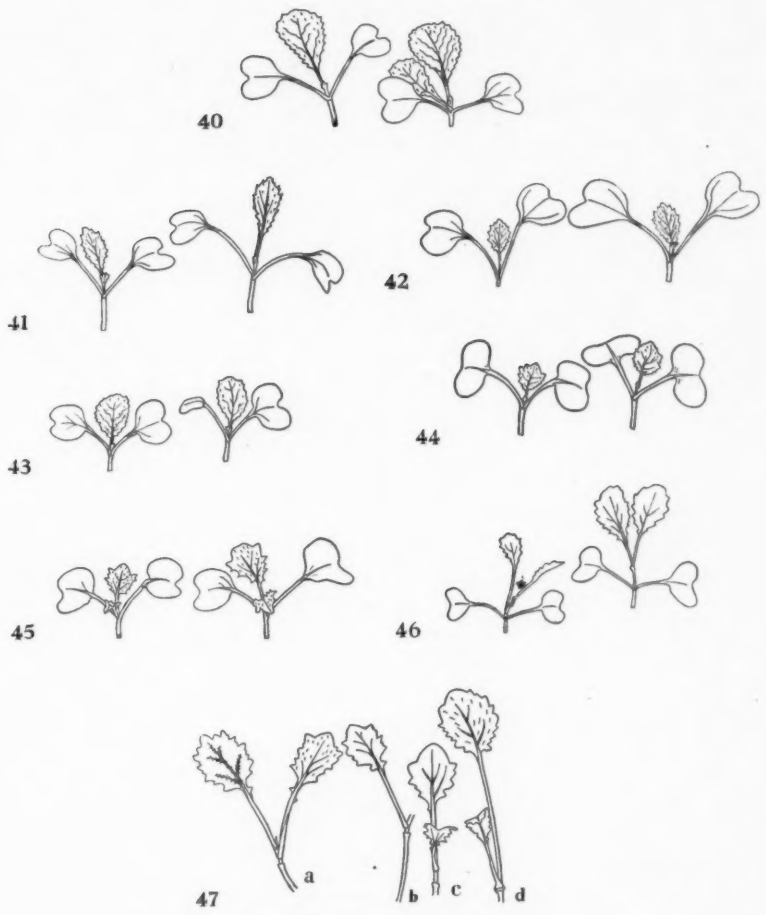


FIG. 40. *B. pekinensis Rupr. Wong bok.*
FIG. 41. *B. pekinensis Rupr. Chihili, Improved Peking.*
FIG. 42. *B. pekinensis Rupr. Chefoo.*
FIG. 43. *B. pekinensis Rupr. Market Pride.*
FIG. 44. *B. Napus L. Colza rape.*
FIG. 45. *B. Napus L. Dwarf Essex rape.*
FIG. 46. *B. Napus L. var. quadrivalvis Hook, fils Thoms.*
FIG. 47, *a*. *B. Napus L. Dwarf Essex rape.*
47, *b*. *B. Napus L. Winter rape.*
47, *c*. *B. Napus L. Colza rape.*
47, *d*. *B. napobrassica (L.) Mill. Rutabaga.*

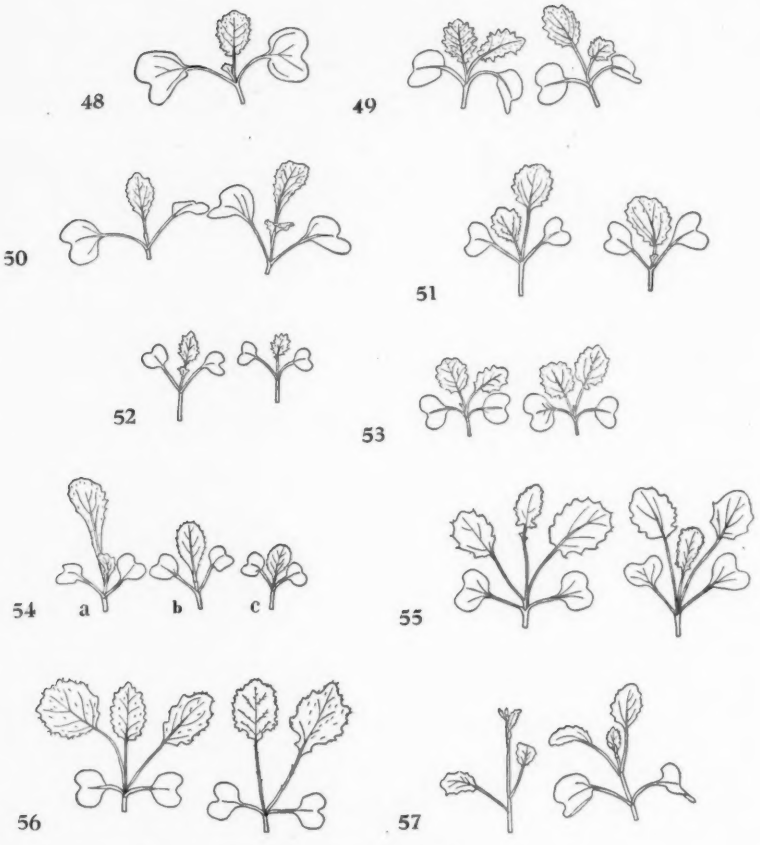


FIG. 48. *B. napobrassica* (L.) *Mill. Rutabaga.*
FIG. 49. *B. Rapa L. var. rapifera Metz. Turnip.*
FIG. 50. *B. Rapa L. Shogoin turnip.*
FIG. 51. *B. Rapa L. Strap leaf turnip.*
FIG. 52. *B. Rapa L. var. septiceps Bailey. Seven-top turnip.*
FIG. 53. *B. campestris L. Wild form.*
FIG. 54 *a*. *B. Rapa L. Bailey. Shogoin turnip.*
54 b. *B. chinensis* (L.) *Bailey. Pak choi.*
54 c. *B. Rapa var. periviridis Bailey. Tendergreen.*
FIG. 55. *B. campestris L. Biennial form.*
FIG. 56. *B. campestris L. Wild form.*
FIG. 57. *B. campestris L. Annual form. Bird rape.*

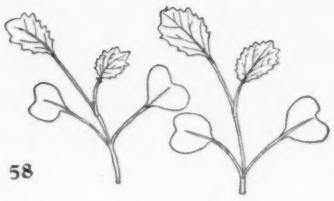

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FIG. 58. *B. campestris L. Annual form. Bird rape.*
FIG. 59. *B. campestris L. Annual form. Bird rape.*
FIG. 60. *B. juncea (L.) Coss. Guy toy.*
FIG. 61. *B. juncea var. crispifolia Bailey. Curled mustard.*
FIG. 62. *B. juncea var. foliosa Bailey. Florida broadleaf.*
FIG. 63. *B. juncea (L.) Coss. Wild form.*
FIG. 64. *B. integrifolia (West) Schulz.*
FIG. 65. *B. arvensis (L.) Rabenh. Wild mustard.*
FIG. 66. *B. nigra (L.) Koch. Black mustard.*
FIG. 67. *B. alba (L.) Rabenh. White mustard.*

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